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The selective group mGlu2/3 receptor agonist LY379268 suppresses REM sleep and fast EEG in the rat

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Abstract

Studies of ionotropic receptors indicate that glutamate (Glu) neurotransmission plays a role in sleep. Here, we show for the first time that metabotropic 2/3 Glu (mGlu2/3) receptors play an active or permissive role in the control of REM sleep. The potent, selective, and systemically active mGlu2/3 receptor agonist LY379268 was administered systemically in doses of 1.0 and 0.25 mg/kg sc. The drug produced a dose-dependent suppression of rapid eye movement (REM) sleep and fast (10–50 Hz) EEG in non-rapid eye movement (NREM) sleep. The 1.0-mg/kg effect on REM sleep was remarkably powerful: REM sleep was totally suppressed in the 6-h postinjection and reduced by 80% in the next 6 h. NREM duration was unchanged during the REM suppression in spite of the strong and unusual depression of EEG power in fast NREM frequencies. These sleep and EEG effects were unaccompanied by motor or behavioral abnormalities. We hypothesize that the REM and the fast EEG suppression were both caused by a depression of brain arousal levels by LY379268. If correct, depressing arousal by reducing excitatory neurotransmission with an mGlu2/3 receptor agonist produces electrophysiological effects that differ drastically from those produced by depressing arousal by enhancing neural inhibition with GABAergic drugs. This different approach to modifying the excitation/inhibition balance in the brain might yield novel therapeutic actions.

Keywords: Sleep; EEG; FFT; REM; Glutamate; Metabotropic

1. Introduction

Mammalian sleep consists of two qualitatively different brain states, non-rapid eye movement (NREM) and rapid eye movement (REM) sleep. Considering the global nature of these states, it would not be surprising to find multiple neurotransmitter systems involved in their regulation. Most research on REM sleep has focused on cholinergic control systems (cf. Jones, 1991; Thakkar et al., 1998; Vazquez and Baghdoyan, 2001). However, the recent discovery that hypocretin/orexin neurons are reduced in narcolepsy (Kilduff and Peyron, 2000; Nishino et al., 2000; Thannickal et al., 2000) now implicates peptidergic neurotransmission in REM sleep.

Research on NREM sleep regulation has long focused on serotonergic neurotransmission (cf. Jouvet, 1999). However, studies over the past decade suggest that adenosine (Benington et al., 1995; Porkka-Heiskanen et al., 1997) and immune factors (Krueger, 1990) may also be involved. Our laboratory has been investigating the role of glutamate (Glu) transmission in sleep regulation. Our initial studies were based on the hypothesis that an experimental increase in the metabolic rate of plastic brain systems would increase NREM delta. NREM delta is thought to be a correlate of a homeostatic process by which sleep reverses the effects of plastic neuronal activity during waking (Feinberg, 1974). We tested this hypothesis by administering systemically ketamine and MK-801, noncompetitive antagonists of the NMDA receptor that are known to produce strong and reliable increases in the metabolic rate of limbic structures (Crosby et al., 1982; Kurumaji et al., 1989). Following the initial period of drug intoxication during which limbic metabolism is increased, both drugs massively increased the high-amplitude, slow-wave (delta) EEG of NREM sleep (Feinberg and Campbell, 1993; Campbell and Feinberg, 1996). Our recent finding that CPPene does not stimulate NREM delta (Campbell et al., 2002) is consistent with but does not prove that the delta stimulation is a response to increased limbic metabolism.

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CPPene is a competitive NMDA receptor antagonist but does not increase limbic metabolism when administered systemically (Boddeke et al., 1992).

Other data suggest that Glu neurotransmission also plays a more direct role in sleep regulation. Thus, perturbations of ionotropic Glu receptors that activate cholinergic systems can modify REM sleep (Manfridi and Mancia, 1996; Cape and Jones, 2000). In addition Glu release was increased during REM sleep in the rostromedial medulla (Kodama et al., 1998), a possible REM control center. Azuma et al. (1996) showed that Glu levels in the medial preoptic area of rats vary with vigilance state, increasing across waking and peaking at the onset of NREM sleep. Glu is also the main neurotransmitter for the retinohypothalamic tract (for a review, see Ebling, 1996).

We thought it of interest to investigate the role of mGlu receptors in sleep for two reasons. First, sleep states are relatively protracted (cf. Evarts, 1967). It therefore seemed likely that, in addition to the relatively short-acting ionotropic receptors, longer-acting receptor systems that control metabolic processes through second messengers would be involved. Second, there is a compelling clinical need for better hypnotic and anxiolytic agents. It is at least logically possible that sleep and sedation could more effectively be promoted by drugs that reduce excitatory neurotransmission than by the classical drugs that enhance neural inhibition at the $GABA_A$–benzodiazepine complex. These considerations pointed to LY379268 as an interesting candidate for sleep studies. LY379268 is a very potent and selective mGlu2/3 receptor agonist (Monn et al., 1999; Schoepp et al., 1999) that is structurally related to LY354740, an earlier mGlu2/3 agonist compound which is anxiolytic in animal studies (Helton et al., 1998). In certain other animal models of psychiatric and neurological disorders (e.g., psychosis and neuroprotection), LY379268 is even more potent and effective than LY354740. Here, we examined doses of LY379268 that are active in these models for their effects on sleep and EEG in rats.

2. Methods

2.1. Animals

Twelve male Sprague–Dawley rats (Simonsen Labs.), 300–350 g at the start of the experiment.

2.2. Surgery

Under deep pentobarbital anesthesia (65 mg/kg), rats were implanted with flexible wire EMG electrodes in the nuchal muscles, a stainless steel screw ground electrode over the olfactory bulb, and six cortical screw electrodes over the frontal, frontoparietal, and parietal cortices at the following coordinates (mm) relative to the bregma: anterior–posterior (AP) +4.5, lateral (L) +2.5; AP +1.5, L +3.5, AP −1.5, L +2.5; AP −4.5, L +3.5; AP +1.5, L −3.5; AP −1.5, L −2.5. Leads from the electrodes were inserted into a small connector cemented to the rat's skull with dental acrylic. The animals were allowed at least 2 weeks to recover from the surgery after which they were gradually trained to the recording apparatus, a counterbalanced cable, and commutator that allowed the rat free movement about the cage. During training, EEG was recorded from all possible ipsilateral pairs of electrodes, and the three cleanest signals were selected for recording in the experiment. The best of these three signals was used for EEG analysis. All recordings were made with the rats in their home cage in a temperature (20–22 °C)- and light (12:12)-controlled room. The UC Davis Animal Use and Care Advisory Committee approved all procedures.

2.3. EEG recording and analysis

EEG and EMG signals were amplified and filtered with Grass amplifiers. Amplifier filters were set at 0.3 Hz low, 100 Hz high for EEG, and at 3 Hz low and 500 Hz high for EMG. Notch (60 Hz) filters on the amplifiers were disabled at all times as they attenuate amplitudes over a wide frequency range. PASS PLUS (Delta software, St. Louis) digitized the amplified signals at 256 Hz and performed online power spectral analyses with the Fast Fourier Transform (FFT). A 200-μV, 10-Hz sine wave calibrated PASS PLUS before each recording session and the measurements on each channel were scaled to this signal. FFT windows were 4 s Welch tapered windows with 2-s overlap, yielding five windows per 10-s epoch. FFT analysis yields frequency bands that differ from integer values; for example, with these FFT parameters, delta was 1.25–4.25 Hz rather than 1–4 Hz. For simplicity of presentation, integer values are used in this report. Frequency bands were 1 Hz wide for 1–4 Hz, 2 Hz wide for 4–12 Hz, 12–15 Hz, 5 Hz wide for 15–35 Hz, and 35–50 Hz. For statistical analyses, these bands were collapsed into the following a priori determined bands: 1–4, 4–6, 6–10, 10–20, 20–30, and 30–50 Hz.

2.4. Scoring of vigilance states

Computer pattern recognition of vigilance states is still unsatisfactory. Therefore, each 10-s epoch was scored visually as NREM, REM, or wake using an on-screen display of the digitized data. PASS PLUS also allowed the scorer access to plots of period–amplitude analyzed wide band EMG and FFT analyzed delta EEG and rho band EEG power. Rho power (25–30 or 30–35 Hz depending on the animal) in REM sleep is elevated above that in both NREM and waking and so provides an indicator of REM sleep (Campbell and Feinberg, 1993), eliminating the need for a hippocampal (theta) electrode.

The criteria for vigilance states were as follows: Wake—low-amplitude, high-frequency EEG with high EMG activity; NREM—high-amplitude, low-frequency EEG with low

EMG activity; REM—low-amplitude, high-frequency EEG (often dominated by theta) with high rho spectral power and very low EMG activity. A second scorer checked all scoring. Epochs containing artifacts (low-frequency movement artifacts and high-frequency eating artifacts) were excluded from FFT analyses but were included in state durations. The vigilance state scores and computer analyzed EEG databases were linked and FFT power was summed for each frequency band in each hour of each vigilance state.

2.5. Behavioral observations

Each animal's behavior was observed for 2 h postinjection and notations were made in the laboratory notebook.

2.6. Drug, administration, and recording schedule

LY379268, (−)-2-oxa-4-aminobicyclo[3.1.0]hexane-4,6-dicarboxylate, was generously provided in powder form by E. Lilly, It was dissolved in equimolar NaOH prior to each experiment. For each animal, the experiment consisted of three consecutive 24 h recording periods: In the first period, a subcutaneous injection of saline was given at hour 6 of the dark (active) period; in the second 24 h period the rats received either 0.25 or 1.0 mg/kg LY379268 sc (six rats each); and the third 24 h period constituted a no-injection recovery recording. EEG and EMG were recorded and digitized at 256 Hz continuously throughout each 3-day session.

2.7. Statistics

The vigilance state durations and EEG power data were summed for each consecutive 6-h block of the experiment. ANOVA with time and condition (saline vs. drug) as repeated measures and dose as a grouping factor yielded significant interactions between the factors for most of the variables examined. Therefore, to determine the effect of LY379268 at the different time points, we performed the following statistical analyses. *T* tests were performed to compare differences from saline for each measure in each 6-h block for each dose. Because of the large number of tests, alpha was set a priori at .01. When both drug doses differed significantly from saline, an ANOVA with dose as a grouping factor and saline values as covariates (BMDP2V, Dixon et al., 1990) was performed to determine whether the effect of 1.0 mg/kg was significantly larger than that of 0.25 mg/kg. The a priori alpha level for this ANOVA was .05.

3. Results

3.1. REM sleep durations

Fig. 1 shows that 1.0 mg/kg LY379268 significantly ($t_{10} = 10.7$, $P = .0001$) reduced minutes of REM sleep by 100% (i.e., to zero) in the 6 h after injection and by over 80% ($t_{10} = 7.6$, $P = .0006$) in the next 6 h. REM sleep was still depressed (but nonsignificantly) in the last 6 h of the injection day. REM durations in the four 6-h blocks of the recovery day did not differ from the corresponding baseline (saline) time periods. The REM-suppressive effects of 0.25 mg/kg LY379268 were qualitatively similar to but smaller and shorter lasting (Fig. 1) than those of the 1.0 mg/kg dose. ANOVAs performed for the first 6-h period following injection of 1.0 and 0.25 mg/kg revealed highly significant dose dependence for REM sleep suppression [$F(1,9) = 50.2$, $P < .0001$]. Beginning in the time block 19 h after injection (i.e., on the recovery day), REM sleep was consistently elevated above baseline by the 0.25-mg/kg dose, but this effect was significant for only the 37–42-h period. For the 1.0-mg/kg dose, REM sleep was nonsignificantly above baseline in two of the four time blocks on the recovery day. One question that arises from the increased REM on the recovery day is whether its amount approximated the REM lost during suppression. An examination of the absolute values showed that this was not the case; after 1.0 mg/kg, 46 min of REM were lost on the injection day vs. 2 min above baseline on the recovery day. An independent question of theoretical interest (see below) is whether NREM duration was increased when REM was suppressed. Although not statistically significant, NREM increased above baseline in the first 6-h period but was slightly below baseline in the second.

3.2. NREM sleep durations

Fig. 1 shows that NREM durations were not significantly changed during the two 6-h periods of strong REM suppression immediately following injection. However, there were some small but significant reductions in NREM durations in the first two 6-h blocks on the recovery day. In the last 6-h period of the recovery day following 0.25 mg/kg, NREM duration was significantly above that in the corresponding baseline period.

3.3. Wake durations

Time awake varied inversely with the changes in NREM and REM sleep durations but Fig. 1 shows that these effects only occasionally reached significance.

3.4. FFT analyses of EEG frequencies

These results are shown in Fig. 2 for the first four 6-h time blocks following drug injection. Data are presented only for NREM and wake since the suppression of REM sleep in the first two time blocks following injection precluded its effective statistical analysis.

3.5. Non-rapid eye movement

Fig. 2 shows that 1.0 mg/kg LY379268 significantly and rather uniformly suppressed fast (10–50 Hz) EEG power by almost 50% in the 6 h following injection. The smaller suppression in the second 6-h time block was also significant for 10–50 Hz frequencies; in addition, power was also significantly depressed in two of the slower (<10 Hz) frequency bands in this period. Several of these effects remained significant in the third 6-h time period but power in all EEG frequencies was back to saline levels in the fourth time block. The changes in NREM EEG following 0.25 mg/kg LY379268 paralleled but were smaller and shorter lasting than those following 1.0 mg/kg. The 0.25-mg/kg dose significantly increased NREM delta power in the first time block. Delta was also increased by the 1.0-mg/kg dose, but this larger effect was not statistically significant due to high variability. Suppression of NREM EEG power in EEG frequencies >10 Hz in the first 6-h time block following injection showed significant dose dependence for 10–20 and 20–30 Hz and approached significance ($P<.06$) for 30–50 Hz.

3.6. Waking

Suppression of fast EEG was also present in waking but the effect was smaller than in NREM sleep (Fig. 2). In the first 6-h time block following injection of 1.0 mg/kg, power in all frequency bands above 6 Hz was significantly depressed. By the third time block, only the suppression of 30–50 Hz power was still significant. In the fourth time block, power in all frequency bands was back at baseline levels. Since delta waves in the waking EEG are an important sign of toxicity, it is worth noting that there was no significant increase in waking delta with either LY379268 dose. In view of the theoretical importance attributed to theta frequencies in the rat (Greenstein et al., 1988; Vinogradova, 1995), it is interesting that the 1.0-mg/kg dose seemed to produce a more marked depression of waking theta as compared to neighboring frequencies. The effects of the 0.25-mg/kg dose on waking EEG again paralleled those of the larger dose but were smaller and shorter lasting. With the exception of 6–10 Hz power ($P=.016$), none of the ANOVAs for dose dependence of LY379268 effects on waking EEG frequency bands approached significance.

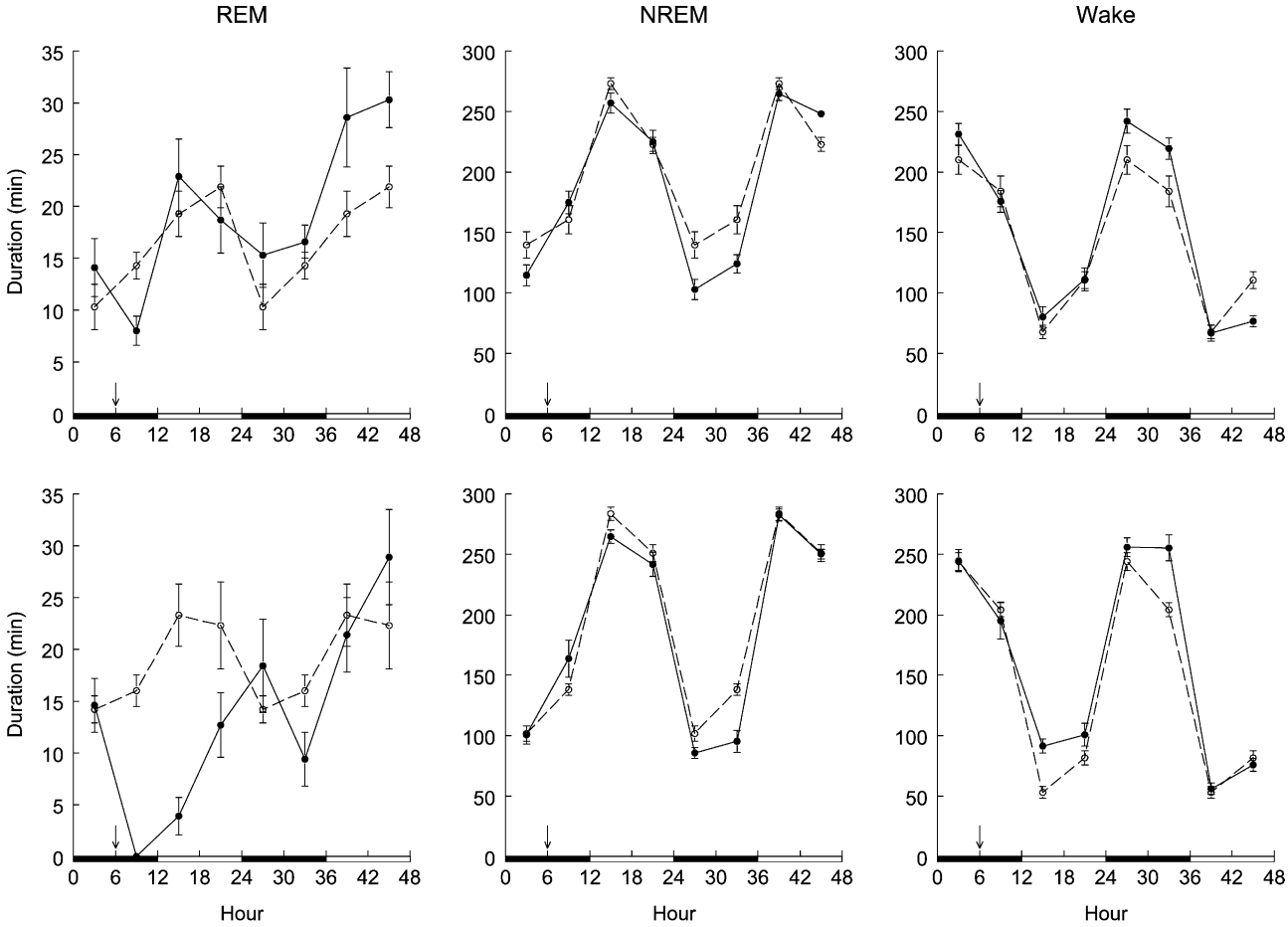

Fig. 1. LY379268 specifically and dose dependently suppressed REM sleep. Effects of 0.25 mg/kg (top row) and 1.0 mg/kg (bottom row) LY379268 on vigilance state durations. 24-h saline data are double plotted. Injected at hour 6 (↓) of the dark period, LY379268 (filled circles, solid line) immediately and dose dependently decreased REM duration relative to saline (open circles, dashed line). Both doses induced smaller delayed effects on NREM and waking.

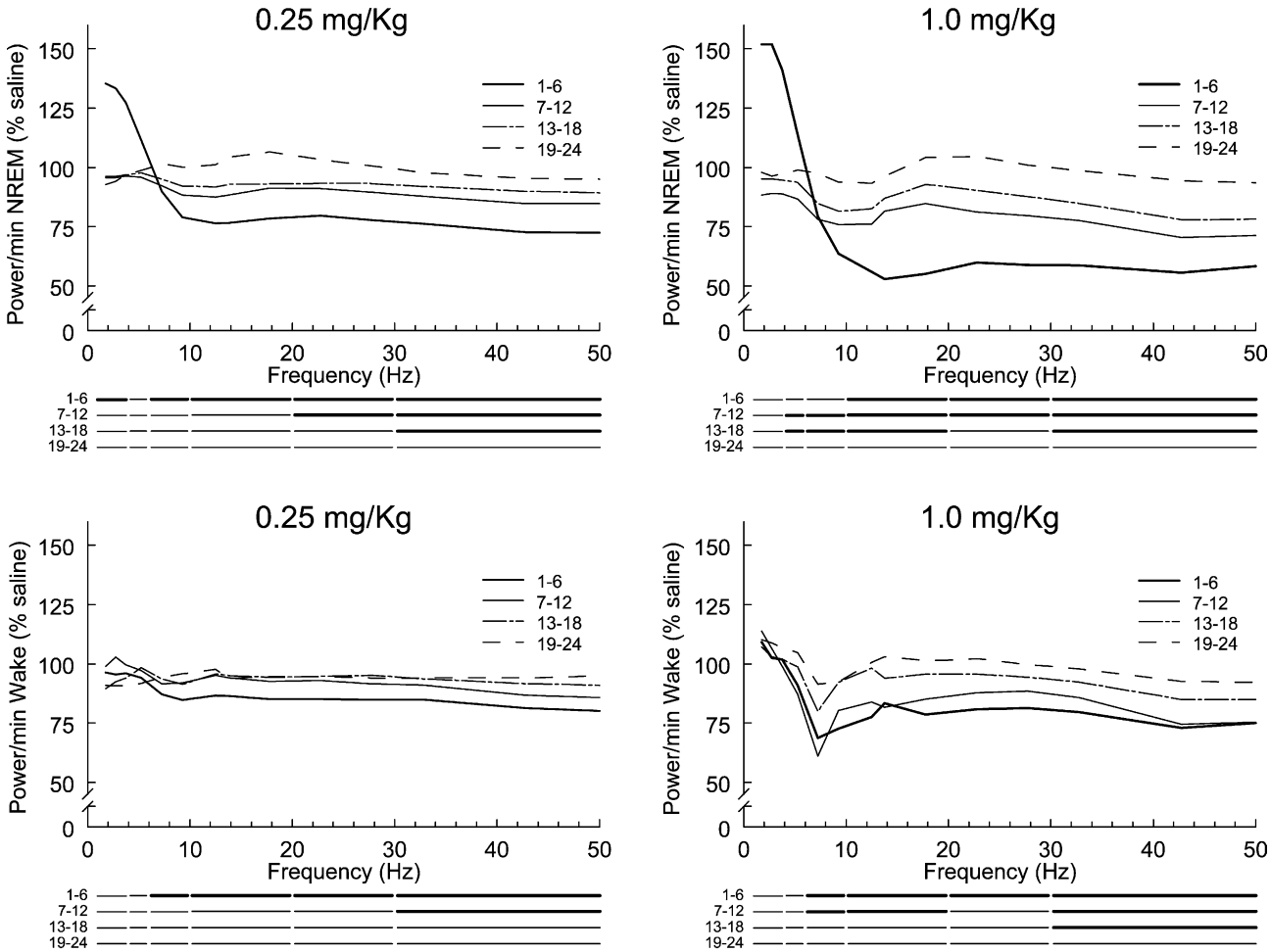

Fig. 2. LY379268 suppressed high-frequency EEG in NREM sleep and waking. Effects of 0.25 (six rats) or 1.0 mg/kg (six rats) LY379268 on EEG spectral power in NREM sleep (top row) and waking (bottom row) in the four successive 6-h periods following injection. Values are expressed as percent of saline control. The frequency bands examined are indicated by bars under the abscissae. Bold bars indicate power in that frequency band was significantly different from saline at $P<.01$ by paired t test. ANOVA for dose dependence was performed for the first 6-h period postinjection. Dose dependence was significant ($P<.01$) in NREM for 10–20 and 20–30 Hz bands and showed a trend ($P=.06$) for 30–50 Hz. The EEG effects in wake were smaller than in NREM. Dose dependence in the 6-h postinjection was significant ($P=.02$) only for 6–10 Hz. These analyses were conducted on data normalized for individual differences in EEG power by expressing each animal's values as a percent of its total power summed for all vigilance states across the 24 h of the saline day.

3.7. Rapid eye movement

The effects of the 0.25-mg/kg dose of LY379268 on REM EEG (not shown) resembled those in waking. As mentioned above, effects of the 1.0-mg/kg dose on REM EEG could not be tested statistically because of the REM suppression.

3.8. Behavioral observations

The 1.0-mg/kg dose of LY379268 produced a distinct reduction in the animals' overall activity level in the 2 h following injection. This suggests a sedative effect. However, in spite of the profound sleep and EEG changes produced by LY379268, we did not observe any pathological motor changes. We were particularly vigilant for ataxia and motor stereotypies (head wagging, circling) but found no such behaviors.

4. Discussion

These data demonstrate for the first time that selective activation of mGlu2/3 receptors profoundly and specifically suppresses REM sleep in the rat. Thus, they add mGlu receptors to the neurotransmitter systems that play either an active or permissive role for REM sleep. During the 6-h period of total REM suppression following 1.0 mg/kg, NREM fast EEG was strongly depressed but NREM durations were unaffected. There were small but statistically significant reductions of NREM durations in two 6-h time

blocks on the recovery day following 1.0 mg/kg LY379268. While it is possible that these were effects of low residual drug concentrations, we think it more likely that they are responses to the previous sleep perturbations.

It is striking that the profound REM and EEG effects were unaccompanied by any pathological motor behaviors. While we did not administer motor tests because we did not wish to alter spontaneous sleep–wake rhythms, other studies have shown that doses of LY379268 of up to 3 mg/kg do not impair rotorod performance in the 8 h following administration (Cartmell et al., 2000). We did observe a reduction in overall motor activity following the 1.0-mg/kg dose. These qualitative observations are consistent with the significant reduction in motor activity documented by Cartmell et al. (1999) using an activity monitor. Toxic drug effects are often manifested by increased slow waves during waking. We recorded no waking EEG slowing following LY379268.

We think it probable that LY379268's suppression of REM sleep and fast EEG are related effects and that both are caused by a functional depression of central arousal systems. The combination of increased slow and decreased fast EEG generally indicates lowered brain arousal. Decreased brain arousal could be expected to reduce REM sleep because REM is a state of heightened cortical arousal (''activation'') within sleep (cf. (Jones, 1991; Thakkar et al., 1998)). While there is considerable evidence that mGlu2/3 agonists modulate neuronal excitability (cf. Schoepp, 2001), it is not known whether they act directly on cholinergic neurons in the reticular activating system. However, it has been shown that perturbations of Group I and Group II mGluRs can modulate striatal acetylcholine release. In addition, the heavy concentration of mGluRs in the reticular nucleus of the thalamus raises the possibility that they control central excitability levels. It is also pertinent that this nucleus plays a major role in controlling cortical electrophysiology during sleep (Steriade et al., 1993).

One possible explanation of how depressing brain arousal would reduce REM sleep is provided by the one-stimulus model (Feinberg and March, 1988, 1995) of NREM–REM interaction. In this model, NREM and REM are different stages in the brain's response to a single, recurrent, inhibitory stimulus. This stimulus first induces NREM sleep, causing the EEG to slow, depressing the firing rate of neurons in most brain structures, reducing cerebral metabolic rate, and shutting down memory acquisition systems. This constellation of brain events provides the biological context in which the putative homeostatic processes of NREM sleep can take place. When the strength of the inhibitory stimulus that initiated NREM sleep falls below a critical level of brain arousal, escape from inhibition occurs. This disinhibited state is REM sleep and it is characterized by intense irregular firing (Evarts, 1967) in many neuronal systems, especially those that are ''hard-wired''.

The one-stimulus model can account for a wide range of NREM and REM phenomena (Feinberg and March, 1995). In the context of the present results, it is relevant that increasing arousal level with chemical (Datta and Siwek, 1997) or sensory (Arankowsly-Sandoval et al., 1987) stimulation during sleep increases the amount of REM. According to the one-stimulus model, REM sleep is suppressed by LY379268 because the drug depresses central arousal systems sufficiently to prevent the brain from reaching the excitation threshold needed to escape from the inhibitory NREM stimulus. This model would predict that reducing REM by decreasing arousal should increase the amount of NREM sleep since escape from its inhibitory effects would be delayed. A nonsignificant increase in absolute minutes of NREM was observed in the first 6-h period of strong REM suppression but not in the second period. The latter finding could indicate deficiencies in the model. Alternatively, it may be the result of a cascade of interacting—and at present unpredictable—responses to the initial REM suppression.

One alternative to the arousal explanation for LY379268's sleep and EEG effects may be related to the fact that mGlu2/3 agonists suppress enhanced excitations via pre- and possibly postsynaptic mechanisms (Anwyl, 1999). These effects of LY379268 on synaptic excitations (via cAMP?) or downregulation of cerebral metabolism could conceivably render neurons incapable of the rapid oscillations of membrane potential required to produce EEG fast waves. However, there is fairly strong evidence against the possibility that LY379268 has produced a widespread reduction of cerebral metabolism sufficient to produce such pervasive EEG effects. First, direct measurement of waking brain metabolism with 2-deoxyglucose reveals that LY379268 causes a slight (nonsignificant) overall increase rather than reduction of glucose uptake (Lam et al., 1999). However, it must be acknowledged that these findings are not conclusive because glucose uptake during sleep following LY379268 has not yet been measured.

A second argument against a general metabolic depression as the cause of the EEG suppression is provided by the findings of Popoli et al. (1999). They microinjected nanomolar quantities of either L-CCG-1 ((2*S*,1′*S*,2′*S*)-2-(2′-carboxycyclopropyl)glycine), a Group II agonist that is also active at Group III receptors (Schoepp et al., 1999) or the Group III agonist L-AP4 (L-2-amino-4-phosphonobutyrate) into the nucleus accumbens of rats and recorded waking EEG for 1 h. These circumscribed and minute injections of a Group II/III and a Group III metabotropic receptor agonist elicited responses in the cortical EEG (delta stimulation and fast EEG suppression) closely resembling those found here with systemic LY379268. Popoli et al. also noted that their EEG findings suggest that L-CCG-1 and L-AP4 depressed arousal. They did not record sleep and it is possible that their accumbens microinjections also suppressed REM sleep.

Current thinking holds that mGlu2/3 agonists normally function to counteract overactivity in Glu systems (Cartmell and Schoepp, 2000). Administration of these drugs to animals in the absence of overactivity might therefore shift the overall balance of brain excitation/inhibition. Such

decreases in excitation might produce therapeutic and neurophysiologic effects that differ from those produced by altering the balance by enhancing inhibition. This possibility is supported by the sleep and EEG findings here. In striking contrast to the sleep and EEG effects of LY379268, clinically used hypnotics that enhance inhibition at the $GABA_A$–benzodiazepine receptor complex suppress slow (delta) and stimulate fast EEG frequencies (cf. Gaillard et al., 1973; Johnson et al., 1979; Borbely et al., 1985; Feinberg et al., 2001). While the GABAergic drugs produce some REM suppression, the effect is modest compared to that of LY379268. An additional difference is that GABAergic drugs depress waking cerebral metabolism (Eintei et al., 1999) but LY379268 does not (Lam et al., 1999). The finding here that LY379268 does not increase total sleep time also supports the behavioral observations that its anxiolytic effects do not depend on the kind of sedation produced by GABAergic drugs. While it is true that mGlu2/3 agonists can modify GABA release (Cartmell and Schoepp, 2000), this effect is unlikely to have produced the findings here since the sleep and EEG effects of LY379268 are so divergent from those of GABAergic hypnotics. They also differ from the sleep and EEG effects of direct $GABA_A$ agonists such as muscimol, which does not suppress REM or fast EEG activity in NREM (Lancel et al., 1996). Taken together, these data indicate that LY379268 reduces central arousal levels by different neurophysiological mechanisms from those activated by the GABAergic modulators.

Clinically, the strong REM suppression by LY379268 raises the possibility that it would have antidepressant effects. Most, but not all, clinically effective antidepressants suppress REM sleep and produce REM rebound on withdrawal (Vogel, 1983). REM suppression is most pronounced with monoamine oxidase inhibitors (MAOIs) (cf. Akindele et al., 1970; Bowers and Kupfer, 1971), which are probably the most effective drugs for severe or refractory depression. The suppression of REM sleep by LY379268 appears to be at least as powerful as that of the MAOIs.

Finally, the remarkable suppression of REM sleep and 10–50 Hz EEG frequencies by LY379268 could be used to investigate some of the functions attributed to these brain states. Ballard et al. (2001) have shown that a closely related mGlu2/3 agonist LY354740 impairs performance of rats on memory and spatial learning tasks. These effects might be mediated by either the REM or the fast EEG suppression. We have unpublished evidence showing that these effects can be dissociated pharmacologically. It should therefore be possible to test whether the impairments found by Ballard et al. are dependent upon either the REM or fast EEG suppression, or both. In addition, LY379268 could be used to test the hypothesis that REM sleep is necessary for memory consolidation (cf. Hennevin et al., 1995) and whether suppression of gamma (30–50 Hz) EEG impairs synchronization of distributed neural activity (cf. Knyazeva et al., 1999). While doses of LY379268 that suppress REM and fast EEG reduce the level of motor activity, they do not cause ataxia or pathological motor behaviors, contributing to the feasibility of behavioral studies.

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