Psychopharmacology (2009) 205:93–106
DOI 10.1007/s00213-009-1519-2

ORIGINAL INVESTIGATION

Influence of the novel antidepressant and melatonin agonist/serotonin2C receptor antagonist, agomelatine, on the rat sleep–wake cycle architecture

Amandine Descamps · Colette Rousset · Mark Millan · Michael Spedding · Philippe Delagrange · Raymond Cespuglio

Received: 7 November 2008 / Accepted: 13 March 2009 / Published online: 16 April 2009

Abstract
Rationale The novel antidepressant, agomelatine, behaves as an agonist at melatonin MT_1 and MT_2 receptors and as an antagonist at serotonin $(5\text{-HT})_{2C}$ receptors. In animal models and clinical trials, agomelatine displays antidepressant properties and re-synchronizes disrupted circadian rhythms.
Objectives The objectives of this study were to compare the influence of agomelatine upon sleep–wake states to the selective melatonin agonists, melatonin and ramelteon, and to the selective 5-HT_{2C} receptor antagonist, S32006.
Methods Rats were administered with vehicle, agomelatine, ramelteon, melatonin, or S32006, at the onset of either dark or light periods. Polygraphic recordings were performed and changes determined over 24 h, i.e., number and duration of sleep–wake episodes, latencies to rapid eye movement (REM) and slow-wave (SWS) sleep, power band spectra of the electroencephalogram (EEG), and circadian changes.
Results Administered at light phase onset, no changes were induced by agomelatine. In contrast, administered shortly before dark phase, agomelatine (10 and 40 mg/kg, per os) enhanced duration of REM and SWS sleep and decreased wake state for 3 h. Melatonin (10 mg/kg, per os) induced a transient enhancement in REM sleep followed by a reduction in REM and SWS sleep and an increase in waking. Ramelteon (10 mg/kg, per os) provoked a transient increase in REM sleep. Finally, S32006 (10 mg/kg, intraperitoneally), administered at dark phase onset, mimicked the increased SWS provoked by agomelatine, yet diminished REM sleep.
Conclusions Agomelatine possesses a distinctive EEG profile compared with melatonin, ramelteon, and S32006, possibly reflecting co-joint agonist and antagonist properties at MT_1/MT_2 and 5-HT_{2C} receptors, respectively.

Keywords Agomelatine · Rat · Melatonin · Sleep · Antidepressant · Polysomnography · S32006 · Ramelteon · Wake

A. Descamps (✉) · C. Rousset · R. Cespuglio
Claude Bernard University,
Lyon EA4170, France
e-mail: amandine.lefebvre@sante.univ-lyon1.fr

M. Millan
Department of Psychopharmacology,
Institut de Recherches Internationales Servier,
Neuilly-sur-Seine, France

M. Spedding · P. Delagrange
Experimental Sciences Department,
Institut de Recherches Internationales Servier,
Neuilly-sur-Seine, France

Introduction

Agomelatine, a novel approach for the treatment of major depression (Kennedy and Emsley 2006; Zupancic and Guilleminault 2006; Olié and Kasper 2007), possesses a unique mechanism of action in that it behaves as an agonist at melatonin MT_1 and MT_2 receptors and as an antagonist at 5-HT_{2C} receptors. Studies conducted in rats have shown that agomelatine, respectively, activates and blocks cerebral populations of melatonin and 5-HT_{2C} receptors (Audinot et al. 2003; Chagraoui et al. 2003; Millan et al. 2003, 2005). Stimulation of MT_1 and MT_2 receptors underlies the ability of agomelatine to resynchronize circadian rhythms of rodents under free-running (constant dark) conditions, and

in procedures involving a delay in the onset of the sleep phase (Armstrong et al. 1993; Redman et al. 1995; Martinet et al. 1996; Van Reeth et al. 1997). These actions are expressed *via* recruitment of melatoninergic mechanisms in the suprachiasmatic nuclei since lesions of this structure abolish the entraining influence of agomelatine upon circadian rhythms (Redman and Francis 1998). The resynchronizing properties of agomelatine have been confirmed in humans. For example, in healthy, elderly men, evening administration of agomelatine led to a phase-shift in circadian rhythms of hormonal release and temperature (Leproult et al. 2005). Such actions are also mimicked in humans by melatonin (Lockley et al. 2000; Rajaratnam et al. 2003).

The ability of agomelatine to enhance extracellular levels of dopamine and noradrenaline in the frontal cortex of conscious rats is mimicked by 5-HT_{2C} antagonists rather than melatonin, and reflects blockade of excitatory 5-HT_{2C} receptors on GABAergic interneurones inhibitory to ascending dopaminergic and adrenergic pathways (Millan et al. 2000, 2003; De Deurwaerdère et al. 2004; Millan 2005). Agomelatine displays antidepressant properties in diverse experimental models in rats (Papp et al. 2003; Bourin et al. 2004; Bertaina-Anglade et al. 2006). In the despair test, 5-HT_{2C} receptor blockade is likely the major mechanism of action (Millan 2005), and melatonin is generally inactive in animal models of depression and does not display antidepressant properties in man (Dalton et al. 2000; Millan 2006). Nonetheless, chronic mild stress is accompanied by a disruption of circadian rhythms (Gorka et al. 1996) and antidepressant actions of agomelatine administered in the morning or evening are mimicked by administration of 5-HT_{2C} antagonists or melatonin, respectively, suggesting that both mechanisms may be involved in the antidepressant actions of agomelatine (Papp et al. 2003; Dekeyne et al. 2008). Further, in the learned helplessness test, antidepressant properties of agomelatine are mimicked neither by a 5-HT_{2C} antagonist nor by melatonin suggesting that a combination of mechanisms may be responsible for its effects (Bertaina-Anglade et al. 2006).

Interest in the influence of agomelatine on sleep is underpinned both by evidence that melatoninergic and serotonergic mechanisms regulate sleep onset, quality and structure (Ursin 2002; Kantor et al. 2002; Cajochen et al. 2003; Millan 2006; Monti and Jantos 2006), and by studies suggesting an interrelationship between depressed states and the sleep–wake cycle. Thus, depressed patients show a decreased latency to the first REM sleep episode together with an enhancement in the amount of REM sleep during the first part of the night. By contrast, the duration of slow-wave sleep (SWS)—or delta sleep—is decreased, and both intermittent wakefulness and early-morning awakening are common (Vogel et al. 1990; Sharpley and Cowen 1995; Ursin 2002; Millan 2006). Interestingly, total sleep deprivation or selective reduction of REM sleep rapidly improves depressed mood (Wirz-Justice and Van den Hoofdakker 1999; Millan 2006). In keeping with these observations, diverse classes of antidepressant suppress REM sleep in both depressed patients and in healthy volunteers, as well as in rodents (Cespuglio et al. 2005), while a rebound elevation in REM sleep has been documented upon withdrawal of SSRIs and SNRIs in patients (Trivedi et al. 1999; Cespuglio et al. 2005). In contrast to the above-mentioned drugs, the atypical agent mirtazapine does not suppress REM sleep despite clinically relevant antidepressant properties (Schittecatte et al. 2002)

In a polysomnographic study conducted in depressed patients, it appears that agomelatine improves sleep continuity. Its administration is, indeed, associated with favorable self-reports of better sleep quality and decreased daytime sleepiness (Quera Salva et al. 2007). In patients with major depressive disorders, perceptions of "getting to sleep" and of "quality of sleep" were significantly superior for agomelatine compared with venlafaxine within 1 week of treatment, and these differences persisted throughout the 6-week trial (Lemoine et al. 2007).

Few data, however, are available concerning the influence of agomelatine on sleep in animals and no rigorous analysis of its effects on the sleep–wake states has as yet been undertaken (Tobler et al. 1994; Grassi-Zucconi et al. 1996). Accordingly, the present study undertook a comprehensive characterization of the influence of agomelatine upon sleep–wake cycles in conscious rats employing an electroencephalographic (EEG) procedure including quantitative, spectral, and circadian analyses. In view of the complexity and onerous nature of these studies, it was necessary to select a limited number of doses of agomelatine for evaluation, and the choice of 10 and 40 mg/kg was based principally upon pharmacological considerations. Thus, these doses are associated with robust anxiolytic, antidepressant, and synchronizing properties (Redman et al. 1995; Millan et al. 2003, 2005; Papp et al. 2003; Zupancic and Guilleminault 2006) and they both recruit melatonin MT_1/MT_2 receptors *and* block 5-HT_{2C} receptors, for which the affinity of agomelatine is lower (Chagraoui et al. 2003, Millan et al. 2003; Zupancic and Guilleminault 2006). Clearly, inasmuch as agomelatine is intended to be used clinically at doses possessing this double mechanism of action, these doses were appropriate for this comparison of its effects to those of melatonin and the novel melatonin MT_1 and MT_2 receptor agonist, ramelteon, shown to induce sleep in various animal models and clinical trials (Kato et al. 2005; Pandi-Perumal et al. 2007). Finally, the effects of agomelatine were also compared to those of S32006, a highly selective antagonist at 5-HT_{2C} receptors which exerts antidepressant and anxiolytic properties in rats (Dekeyne et al. 2008).

Materials and methods

Animals Wistar rats (200–250 g from Charles River, l'Arbresle, France) were employed. All experiments were performed in compliance with the principles of laboratory care according to the relevant decree of the French Agriculture Ministry (No. 03-505).

Surgery Polygraphic electrodes were implanted under chloral hydrate anesthesia (400 mg/kg, i.p.). After full induction of anesthesia, animals were mounted in a stereotaxic frame and their body temperature maintained at 36.5–37°C by use of a homeothermic blanket. Two electrodes (length, 2 mm; diameter, 500 µm; stainless steel and connected to Teflon-insulated wire) were placed into the left and right frontal cortices (2 mm lateral and anterior to the bregma; Paxinos and Watson 1998), and two were also placed into the left and right parietal cortices (2 mm lateral to the midline at the midpoint between bregma and lambda; Paxinos and Watson 1998) for recording of electroencephalograms (EEG). For recording of electromyograms (EMG), three electrodes (active length, 1 mm; diameter, 500 µm, stainless steel and connected to Teflon-insulated iron wires) were inserted between two layers of neck muscles. Following placement, all electrodes were soldered to two miniature five-pin connectors (Sei 3DAY, Lyon, France) and the entire assembly anchored to the rat skull with Super-Bond glue (Sun Medical & Co., Shiga, Japan) and dental acrylic resin (Ivoclar, Lyon, France). Following surgery, animals were housed individually in plastic home cages, which were placed in sound-isolated chambers (ambient temperature, 22±1°C; light–dark cycle 12 h–12 h, water and food ad libitum). They were allowed 1 week for recovery, then connected to recording cables and 1 week was allowed for habituation.

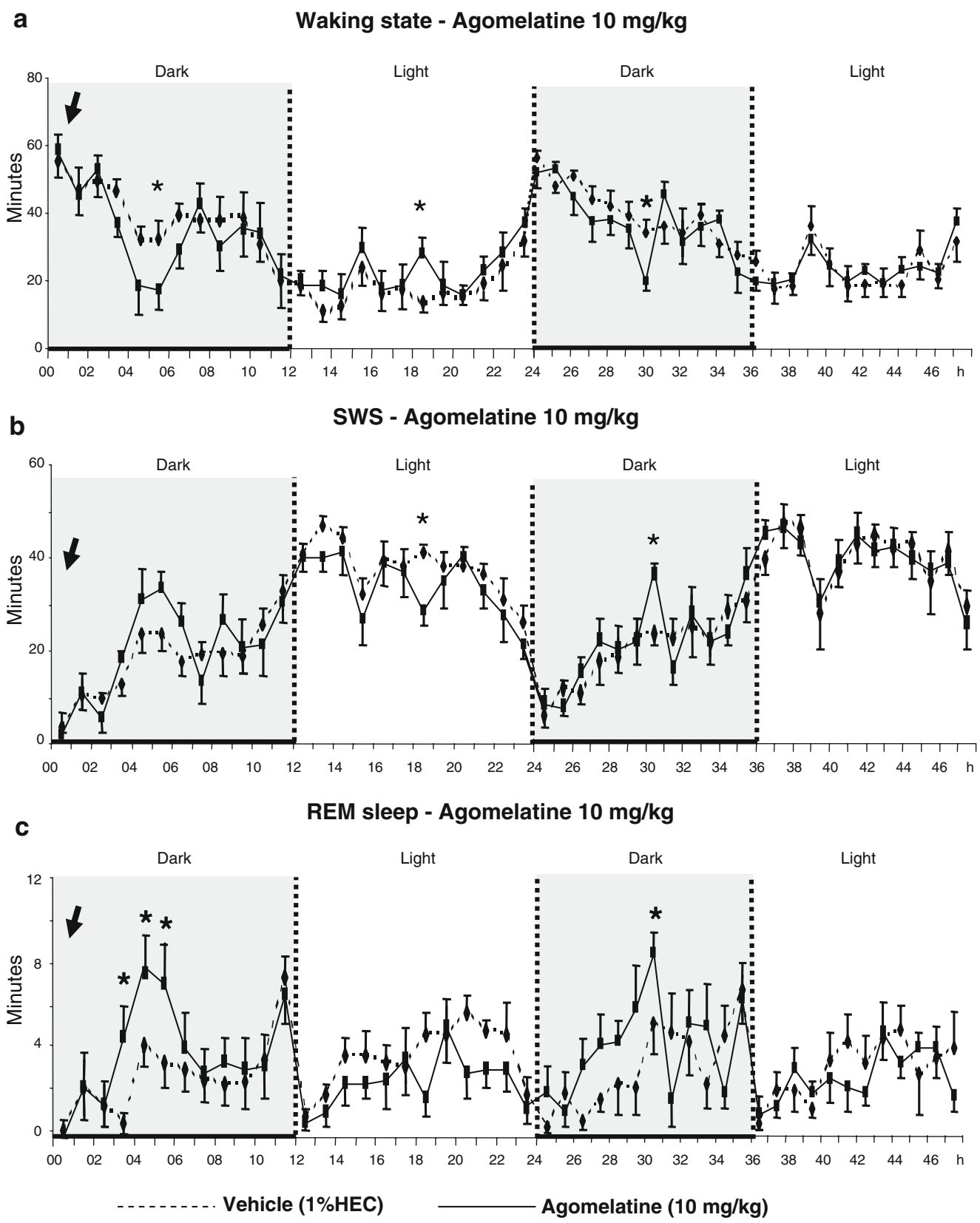
Recordings The procedure employed was essentially that described previously (Clement et al. 2003; Cespuglio et al. 2005). Polygraphic recordings (Embla set-up, Medcare, Iceland) were initiated and continued until stable baselines of sleep–wake states were obtained two full 24 h cycles; each cycle 12 h dark-12 h light. This study was performed in three steps with always one group dosed at light onset and the other one dosed at dark onset. The first was performed with melatonin and its vehicle (water) in the same group (n=6). The second was performed with agomelatine, ramelteon and their vehicle (HEC 1%) administered randomly (Latin square distribution) in the same group (n=6). Lastly the third was performed with S 32006 and its vehicle (HEC 1%, n=12). Visual scoring of digitized EEG and EMG traces (EEG filtering: 0.5–49.9 Hz and EMG: 15–49.9 Hz) was performed over 10 s epochs in order to quantify the number and duration of sleep–wake episodes and the circadian scheduling of sleep–wake states. Power spectra of the EEG (Somnologica software, Medcare, Iceland) were also characterized. To this end, EEG traces sampled at 100 Hz were subjected to fast-Fourier transformation (256 points, computational window, 2.56 s and 50% overlap). Spectra were averaged over 10-s epochs and divided into five adjacent bands (delta, 0.5–4 Hz; theta, 4–8 Hz; alpha, 8–11.5 Hz; sigma, 11.5–14.5 Hz; beta-1, 14.5–18.6 Hz and beta-2, 18.6–30 Hz) and expressed in percent of the total band powers (0–49.9 Hz).

Drugs Agomelatine (S20098: *N*[2-(7-methoxy-1-naphthyl) ethyl] acetamide) was dissolved in hydroxyethylcellulose (HEC) at 1%. Melatonin (5-methoxy-*N*-acetyltryptamine) was dissolved in water. Ramelteon (TAK-375: (*S*)-*N*-[2-(1,6,7,8-tetrahydro-2*H*-indeno-[5,4-*b*]furan-8-yl)-ethyl-propionamide) was dissolved in HEC 1%. S32006 (*N*-pyridin-3-yl-1-2-dihydro-3H-benzo[*e*]indole-3-carboxamide) was dissolved in HEC 1%. All the above substances were administered p.o., except for S32006 which was injected intraperitoneally.

Statistics Following an analysis of variance (ANOVA), comparisons between experimental conditions (first factor: treatments at light or dark onset; second factor: vehicle recordings) were performed by use of Fisher least significant difference tests (LSD). When ANOVA (F values) were significant at $p<0.05$, LSD tests were performed for multiple comparisons. For this purpose, Statgraphics software (Manugistic, Rockville, MD, USA) was used. The influence of agomelatine, ramelteon, melatonin, and S32006 upon waking (W), SWS and REM sleep states was evaluated by ANOVA across individual animals and all drug doses. For hourly comparisons, the second factor was concatenated with time. Finally, the cosine fit analysis was applied as described previously for the circadian rhythm analysis (Cespuglio et al. 2005). In this respect, averaged data were treated in 1-h periods, and the validity of fitting assessed by a Fisher's F-test. For EEG spectra values (expressed as % of total power), drug actions were assessed by ANOVA followed by a post-hoc LSD test (*$p<0.05$).

Results

Effect of agomelatine administered at the onset of the dark period At a dose of 10 mg/kg (day-1, 0–24 h), agomelatine decreased the duration of the waking state compared to vehicle over a 4–7 h time period following administration. The magnitude of this effect reached significance 6 h after administration: −44%, F [47.2]=7.28, $p<0.01$, Fig. 1a. During 12 h of the subsequent light period (12–24 h), only a transient but significant increase was seen in waking 19 h


a
Waking state - Agomelatine 10 mg/kg
Dark
Light
Dark
Light
Minutes
80
60
40
20
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
b
SWS - Agomelatine 10 mg/kg
Dark
Light
Dark
Light
Minutes
60
40
20
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
c
REM sleep - Agomelatine 10 mg/kg
Dark
Light
Dark
Light
Minutes
12
8
4
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
Vehicle (1%HEC)
Agomelatine (10 mg/kg)

◀ **Fig. 1** Influence of agomelatine (10.0 mg/kg, p.o.) upon 48 h measures of waking state (W, **a**), slow-wave sleep (SWS, **b**) and rapid eye movement sleep (REM, **c**) sleep following administration at the onset of the first dark period. Agomelatine was administered (day-1, *arrow*, 10.0 mg/kg per os) at the onset of the dark period and recordings made over two full 24 h (day-1 and day-2) cycles. Dark and light periods are indicated by *shaded* and *non-shaded parts* of the figure, respectively. Number of animals, six for vehicle (HEC, 1% hydrocellulose) and five for agomelatine during day-1, and six for vehicle and agomelatine during day-2. *Ordinate* duration in minutes (each point represents mean duration per 1 h±SEM). *Abscissa* time-scale in hours (h) from "zero" which corresponds to administration of agomelatine. *Asterisks* indicate significance (*$p<0.05$) of agomelatine versus vehicle differences in post-hoc least significant difference (LSD) tests following ANOVA

after administration (Fig. 1a). During day-2 (24–48 h), and particularly during the 12-h dark period (24-36 h), the duration of the waking state decreased and this effect was significant 31 h after administration: −50%, F [47.24]=6.7 $p<0.01$, Fig. 1a. SWS duration varied in an opposite manner. During the 12 h of the subsequent light period, a transient and significant decrease was observed 19 h after administration (Fig. 1b). During day-2 (24–48 h), especially during the 12 h of the dark period (24–36 h), SWS significantly increased 31 h after administration: +54%, F [47.2]=8.7, $p<0.01$, Fig. 1b. The most marked effect of agomelatine was exerted on REM sleep that significantly increased during day-1 and particularly at 4, 5, and 6 h after administration by +500%, +80%, and +108%, respectively, F [47.21]=2.74, $p<0.01$, Fig. 1c. No significant change occurred during the subsequent light period, but during the dark period of day-2, REM sleep exhibited a significant increase 31 h after administration +61%, F [47.2]=2.33, $p<0.01$; Fig. 1c. The latencies to SWS and REM sleep were not significantly modified on either day 1 or 2.

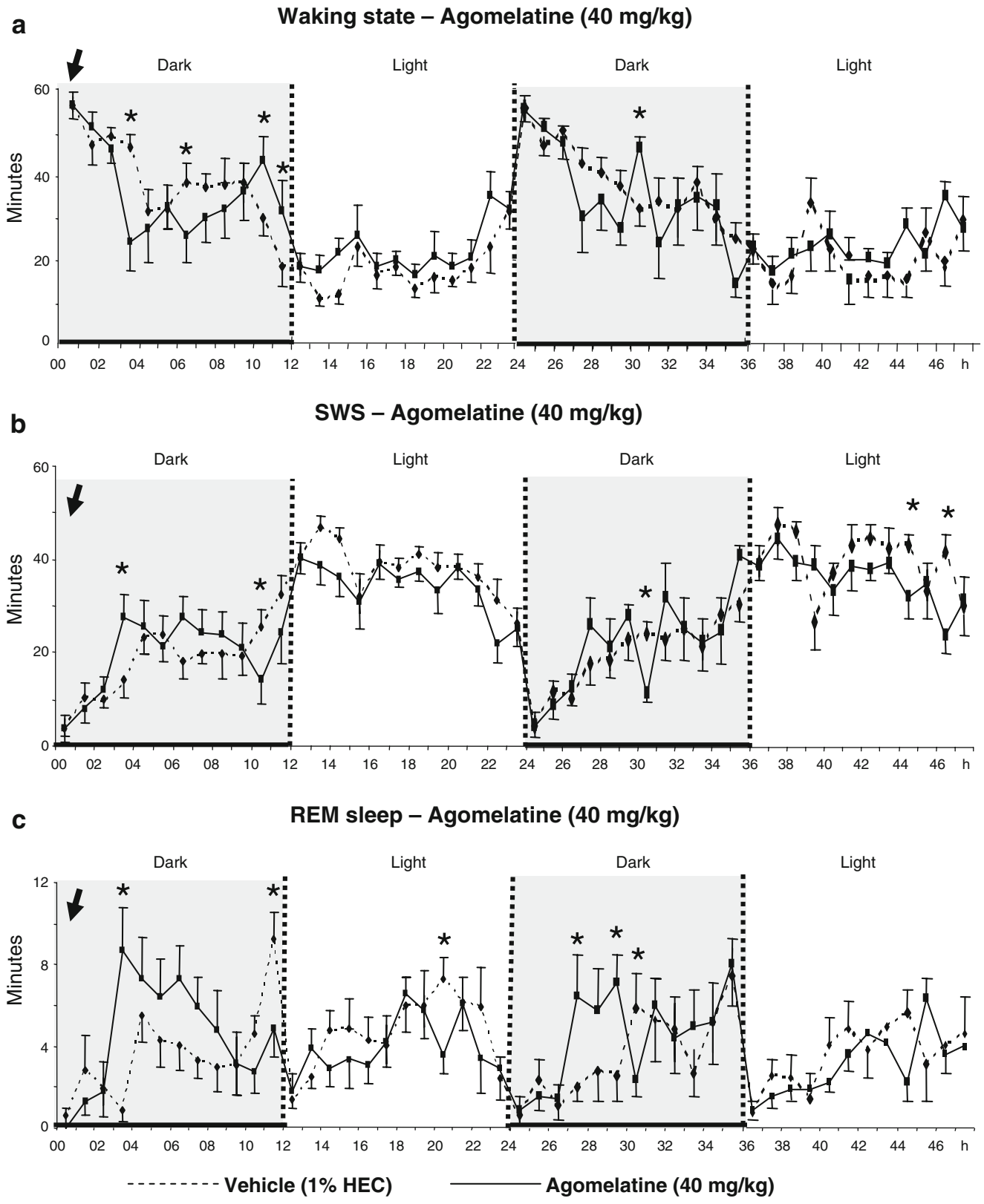
At a dose of 40 mg/kg (day-1, 00–24 h) agomelatine decreased waking state versus vehicle at 4–9 h following administration. The magnitude of this effect reached significance at 4 h with decreases of −48% and −33%, respectively, F [47.24]=7.10, $p<0.01$; Fig. 2a. Just prior to light onset, waking state significantly increased 11 h and 12 h after administration (Fig. 2a). During the 12 h duration of the subsequent light period (12–24 h), no change was seen. During day-2 (24–48 h), and particularly during the 12-h dark period (24–36 h), the waking state was elevated 31 h after administration (Fig. 2a). Duration of SWS varied in an opposite manner. Thus, on day-1, SWS exhibited a significant increase 4 h after administration, +112%, F [47.24]=8.71, $p<0.01$; Fig. 2b. This effect was followed by a transient decrease just prior to the light onset (Fig. 2b). During the 12 h of the subsequent light period, no significant change was apparent. Over day-2 (24–48 h), SWS exhibited only transient and opposite changes (Fig. 2b). The most marked effect of agomelatine was exerted on REM sleep over the 4-9 h time period with a significant increase at 4 h, +903%, F [27.24]=2.87, $p<0.01$, Fig 2c. This effect was followed by a significant decrease just prior to the light onset (Fig. 2c). Except for a transient decrease 21 h after administration (Fig. 2c), no changes were observed for REM sleep during the subsequent light period. However, during the dark period of day-2, REM sleep significantly increased at 28 and 30 h after administration by +248% and +187%, respectively, F [47.24]=2.53, $p<0.01$, Fig. 2c. This effect was followed by transient decrease 31 h after administration (Fig. 2c). Finally, the latencies to SWS and REM sleep were not significantly modified.

For both doses, the cosine components of the sleep wake states were not significantly modified versus vehicle (data not shown) and there was no significant variations in spectral power bands of SWS, REM sleep, and waking state (data not shown).

Effects of agomelatine administered at the onset of the light period Agomelatine (10 or 40 mg/kg) did not induce significant changes in waking state, SWS, and REM sleep versus vehicle throughout the 12 h of the light period (data not shown). This lack of effect contrasts with the changes obtained when administered at the onset of the dark period (Figs. 1 and 2). Latencies to SWS and REM sleep, as well as cosine components and spectral power bands of sleep wake states did not change versus vehicle (data not shown).

Effect of melatonin administered at the onset of the dark period At a dose of 10 mg/kg (day-1) melatonin induced a biphasic effect on waking state. A tendency toward a decrease (non-significant) occurred initially at 3 h (Fig. 3a) followed by an increase of the waking state (5–8 h) which reached significance 7 h after administration, +60%, F [47.43]=5.0, $p<0.01$, Fig. 3a. During the subsequent light period (12–24 h), no change occurred (Fig. 3a). Over the same time period, SWS tended to be facilitated (Fig. 3b) and then was significantly decreased 7 h after administration: −43%, F [47.43]=4.90, $p<0.01$, Fig. 3b. During the subsequent light period (12–24 h), no change occurred (Fig. 3b). REM sleep also exhibited biphasic changes characterized by a significant increase at 3 h (+94%, F [47.43]=2.91, $p<0.01$, Fig. 3c) and a significant decrease at 6 h (−48% F [47.43]=2.91, $p<0.01$) and at 7 h (−64%, F [47.43]=2.91, $p<0.01$, Fig. 3c) after administration. During the subsequent light period (12–24 h), no change was seen (Fig. 3c). Latencies to SWS and REM sleep were not significantly modified, and the cosine components of sleep wake states were also unaffected (data not shown). No significant variations in the spectral band powers of SWS, REM sleep and waking state were seen (data not shown).

Effect of melatonin administered at the onset of the light period At a dose of 10 mg/kg, melatonin did not


a
Waking state – Agomelatine (40 mg/kg)
Dark
Light
Dark
Light
Minutes
60
40
20
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
b
SWS – Agomelatine (40 mg/kg)
Dark
Light
Dark
Light
Minutes
60
40
20
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
c
REM sleep – Agomelatine (40 mg/kg)
Dark
Light
Dark
Light
Minutes
12
8
4
0
00 02 04 06 08 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46 h
Vehicle (1% HEC)
Agomelatine (40 mg/kg)

◀ **Fig. 2** Influence of agomelatine (40.0 mg/kg, p.o.) upon 48 h measures of waking state (W, **a**), slow-wave sleep (SWS, **b**) and rapid eye movement sleep (REM, **c**) sleep following administration at the onset of the first dark period. Agomelatine was administered (day-1, *arrow*, 40.0 mg/kg per os) at the onset of the dark period and recordings made over two full 24 h (day-1 and day-2) cycles. Dark and light periods are indicated by *shaded* and *non-shaded parts* of the figure, respectively. Number of animals, six for vehicle (HEC, hydrocellulose) and five for agomelatine during day-1, and six for vehicle and agomelatine during day-2. *Ordinate* duration in minutes (each point represents mean duration per 1 h±SEM). *Abscissa* time-scale in hours (h) from "zero" which corresponds to administration of agomelatine. *Asterisks* indicate significance (*$p<0.05$) of agomelatine versus vehicle differences in post-hoc least significant difference (LSD) tests following ANOVA. For other abbreviations, see also Fig. 1

significantly affect waking state, SWS, and REM sleep versus vehicle throughout the 12 h of the light period (data not shown).

Effect of ramelteon administered at the onset of the dark period At a dose of 10 mg/kg (day-1), ramelteon induced only a transient decrease in the waking state 4 h after administration, −26%, F [47.2]=10.41, $p<0.01$, Fig. 3d. During the subsequent light period (12–24 h), no change was observed (Fig. 3d). During the same time period, SWS was increased, +65%, F [47.2]=12.8, $p<0.01$, Fig. 3e. In the course of the subsequent light period (12–24 h), no effect was seen (Fig. 3e). REM sleep exhibited a significant increase 4 h after administration, +448%, F [47.24]=2.9, $p<0.01$, Fig. 3f). During the subsequent light period (12–24 h), no change occurred (Fig. 3f). The latencies to SWS and REM sleep were not significantly modified. The cosine components of sleep–wake states were also not significantly modified (data not shown). No significant variations in spectral band powers of SWS, REM sleep, and waking state were seen (data not shown).

Effect of ramelteon administered at the onset of the light period Ramelteon (10 mg/kg) did not significantly change the waking state, SWS, and REM sleep as compared with vehicle throughout the 12 h of the light period (data not shown).

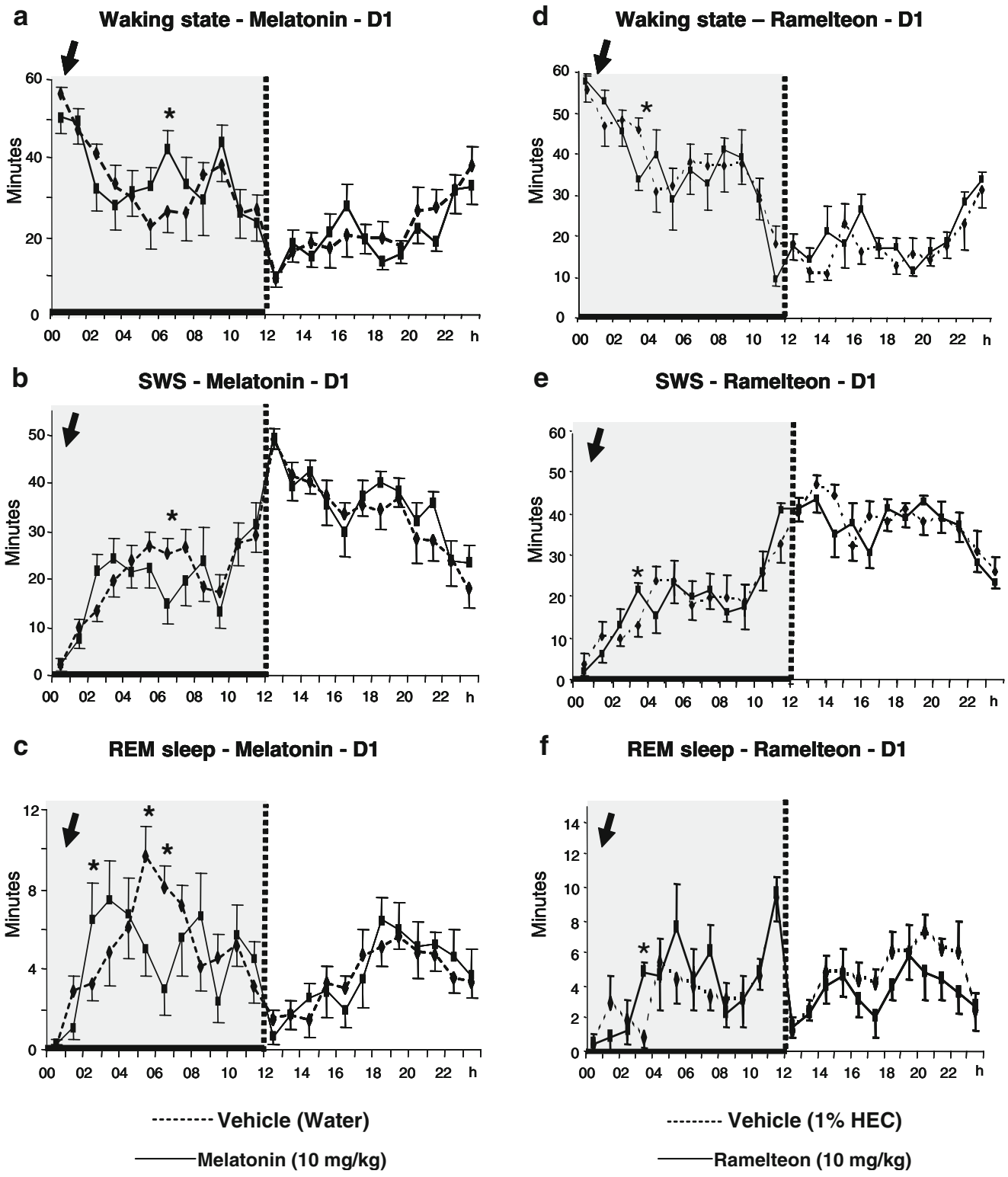
Effect of S32006 administered at the onset of the dark period At a dose of 10 mg/kg (day-1), S32006 exerted a biphasic influence on the waking state. A tendency toward a decrease (non-significant) occurred initially (Fig. 4a) which was followed by a more sustained facilitation (5–8 h) reaching significance 7 h after administration, +33%, F [47.5]=18.17, $p<0.01$, Fig. 4a. During the subsequent light period (12–24 h), a decrease in waking occurred 19–21 h after administration (Fig. 4a). SWS was also influenced in a biphasic manner. A significant increase occurred 4 h after administration followed by a decrease at 7 h, −34.2%, F [47.4]=14.8, $p<0.01$, Fig. 4b. During the subsequent light period (12–24 h), no significant changes were seen (Fig. 4b). REM sleep was significantly decreased 2, 3, 7, and 8 h after administration by −83%, −68%, −74%, and −77%, respectively (F [47.45]=10.12, $p<0.01$, Fig. 4c). The latency to SWS was not significantly modified whereas the latency to REM sleep was prolonged by +140% (vehicle=59.1±4 versus S32006=142.4±17.5; F [1,19]=19.7, $p<0.01$). This change was also reflected in the position of the cosine component of REM (vehicle=9 h 11 versus S32006=8 h 52; curves not shown). There was no significant variation in the spectral band powers of SWS, REM sleep, and waking states (data not shown).

Effect of S32006 administered at the onset of the light period The waking state was decreased 4 h after administration, −39.3%, F [47.5]=7.6, $p<0.05$, Fig. 4d. SWS was successively decreased (1 h, −70.6% and 2 h, −23.8%, F [47.52]=10.26, $p<0.01$; Fig. 4e) and increased (4 h, +35.2%, F [47.52]=10.26, $p<0.01$, Fig. 4e). The latency to SWS was significantly increased (SWS,+53% versus vehicle=39.7±2.7; S32006=60.9±7.14; F [1.22]=7.73, $p<0.05$). This change was reflected in the position of the cosine component (vehicle=15 h 20; S32006=15 h 56; curves not shown). No significant alterations occurred in the spectral band powers of SWS, REM sleep and waking states (data not shown).

Comparison of hourly effects of agomelatine versus the other drugs Administered at dark onset, the most marked increase in SWS was obtained with agomelatine at 40 mg/kg versus ramelteon, melatonin, and S32006 (see Figs. 1, 2, 3, 4). For REM sleep, the greatest increase was also obtained with agomelatine at either 10 at 40 mg/kg versus ramelteon, melatonin, and S32006 (see Figs. 1, 2, 4).

Discussion

Influence of agomelatine as compared to melatonin, ramelteon, and S32006 The present study demonstrates that the melatonin MT_1/MT_2 agonist and 5-HT_{2C} antagonist, agomelatine, administered at 10 or 40 mg/kg, prior to the dark period, induces a consistent pattern of changes in sleep–wake architecture over a period of 3 h (4–7 h following administration). These changes are characterized by increases in both SWS and REM sleep, as well as a decrease in the waking state. These effects disappeared during the subsequent light period of day-1 but were still remarkably apparent (albeit less pronounced) during the next dark period of day-2. Moreover, the specificity and distinctive nature of this profile is underlined by the lack of


a
Waking state - Melatonin - D1
d
Waking state – Ramelteon - D1
b
SWS - Melatonin - D1
e
SWS - Ramelteon - D1
c
REM sleep - Melatonin - D1
f
REM sleep - Ramelteon - D1
Minutes
h
Vehicle (Water)
Melatonin (10 mg/kg)
Vehicle (1% HEC)
Ramelteon (10 mg/kg)

◀ **Fig. 3** Influence of melatonin and ramelteon upon 24 h measures of waking state (W, **a**,**d**), slow-wave sleep (SWS, **b**,**e**) and rapid eye movement sleep (REM, **c**,**f**) sleep following administration at the onset of the dark period. Melatonin (*left panel*) and ramelteon (*right panel*) were administered at the onset of the dark period (*arrow*, 10.0 mg/kg, per os) and their effects monitored throughout the 24 h (day-1, *D1*) following their administration. Dark and light periods are indicated by *shaded* and *non-shaded parts* of the figure, respectively. Number of animals, 12 for vehicle (H_2O) and melatonin, and six for vehicle (*1% HEC*) and ramelteon. *Ordinate* duration in minutes (each point represents mean duration per 1 h±SEM). *Abscissa* time-scale in hours (h), from "zero" which corresponds to the administration of drug. Asterisks indicate significance (*$p<0.05$) of melatonin or ramelteon versus vehicle differences in post-hoc least significant difference (LSD) tests following ANOVA. For other abbreviations, see also Fig. 1

alterations in latencies to SWS and REM sleep, the lack of disruption of circadian (or diurnal) timing, and the preservation of the spectral components of the EEG. Further, agomelatine did not modify the sleep–wake state when administered at the onset of the light period, in agreement with an earlier study of Tobler et al. (1994). Notably, the effects of agomelatine displayed both interesting differences and similarities to the actions of melatonin, the synthetic melatonin agonist, ramelteon, and the novel, selective 5-HT_{2C} antagonist, S32006. The latter substance was the only agent to affect sleep–wake partitioning when given before the light phase suggesting that this action reflects its intrinsic pharmacological actions rather than the chronobiotic properties shared by agomelatine, melatonin, and ramelteon which were inactive when administered at light onset. This "window of sensitivity" for the influence of melatoninergic agents on sleep around dark onset has similarly been reported for their phase-shifting effects in rodents and man (Lewy et al. 1992; Van Reeth et al. 1997; Slotten et al. 2002; Hack et al. 2003). It is worth noting that the effects of agomelatine on sleep though significant at 4–7 h after administration, had subsided by the end of the 12-h dark period. This may be beneficial in permitting the sleep–wake cycle to remain within a physiological temporal range without alterations in the nycthemeral distribution of sleep components. These differences in the duration of effects could not be due to different half-lives which are between 23 min and 2 h in rats for the molecules tested (1 h for S 32006 and 2 h for agomelatine (unpublished data), 23 min for melatonin (Gibbs and Uriend 1981). The half-life for ramelteon in a human is between 0.8 and 1.9 h (Karim et al. 2006).Together with lack of changes in spectral power, these findings concur with observations made in man where agomelatine restored disorganized normal sleep–wake architecture in depressed patients over the first two cycles following its administration (Quera Salva et al. 2007).

Melatonin is considered as the "night" hormone and generally exerts a functional effect mimicking the dark period, corresponding to its actions in the present study in rats, a nocturnal species. Indeed, wake-promoting effects of melatonin have previously been reported both in rats and in hamsters (Huber et al. 1998). Interestingly, its effects herein were biphasic, with a significant increase of REM sleep at 3 h followed by a decrease concomitant with a decrease in SWS and increased waking. A similar biphasic effect of melatonin treatment has been reported in humans. Thus, in a study of healthy men receiving melatonin or placebo for 8 days prior to a 16-h sleep "opportunity", activity (recorded with wrist actigraphy) was reduced in the first half yet increased in the second half of the sleep period. Nonetheless total activity during the sleep and wake periods were not modified (Rajaratnam et al. 2003). Interestingly, despite its lack of effects on total sleep time, melatonin advanced circadian rhythms of endogenous melatonin and cortisol secretion. These observations, consistent with the notion that the phase-shifting effects of melatonin, are more robust than its less comparatively modest effects on various sleep parameters (Hughes et al. 1998; Baskett et al. 2003; Rajaratnam et al. 2003; Wade et al. 2007). Moreover, numerous studies have reported a lack of correlation between endogenous levels of melatonin at night and sleep quality (Hughes et al. 1998; Mahlberg and Kunz 2007). Accordingly, the decreased sleep latency observed after treatment with melatonin or melatonin agonists in insomniacs and patients with "delayed sleep phase syndrome" is probably related to phase-shifting effects rather than an impact on sleep per se (Mundey et al. 2005; Zemlan et al. 2005; Roth et al. 2005).

Possible mechanisms involved in the sleep–wake influence of agomelatine Inasmuch as agomelatine more clearly increased SWS at the dose of 40 versus 10 mg/kg, this tends to support a possible implication of 5-HT_{2C} receptor blockade which is more pronounced at higher doses, in line with the lower affinity of agomelatine for 5-HT_{2C} versus melatonin receptors (Millan et al. 2003; Audinot et al. 2003). In support of this possibility, the 5-HT_{2C} antagonist, S32006, mimicked the SWS reinforcing action of agomelatine upon administration prior to the dark phase, and previous studies have likewise suggested that blockade of 5-HT_{2C} receptors primarily facilitates deep SWS in rats and in man (Dugovic et al. 1989a, b; Sharpley et al. 1994; Kantor et al. 2002; Smith et al. 2002). Nonetheless, it is unclear why agomelatine did not mimic the SWS-enhancing actions of S32006 when given before the light phase. Further, in distinction to agomelatine, which increased REM sleep upon administration before the dark period, S32006 decreased REM sleep, a finding corresponding to studies with other 5-HT_{2C} antagonists (Smith et al. 2002; Monti and Jantos 2006).

The contrasting actions of S32006 when given before the light versus dark periods are intriguing and might reflect

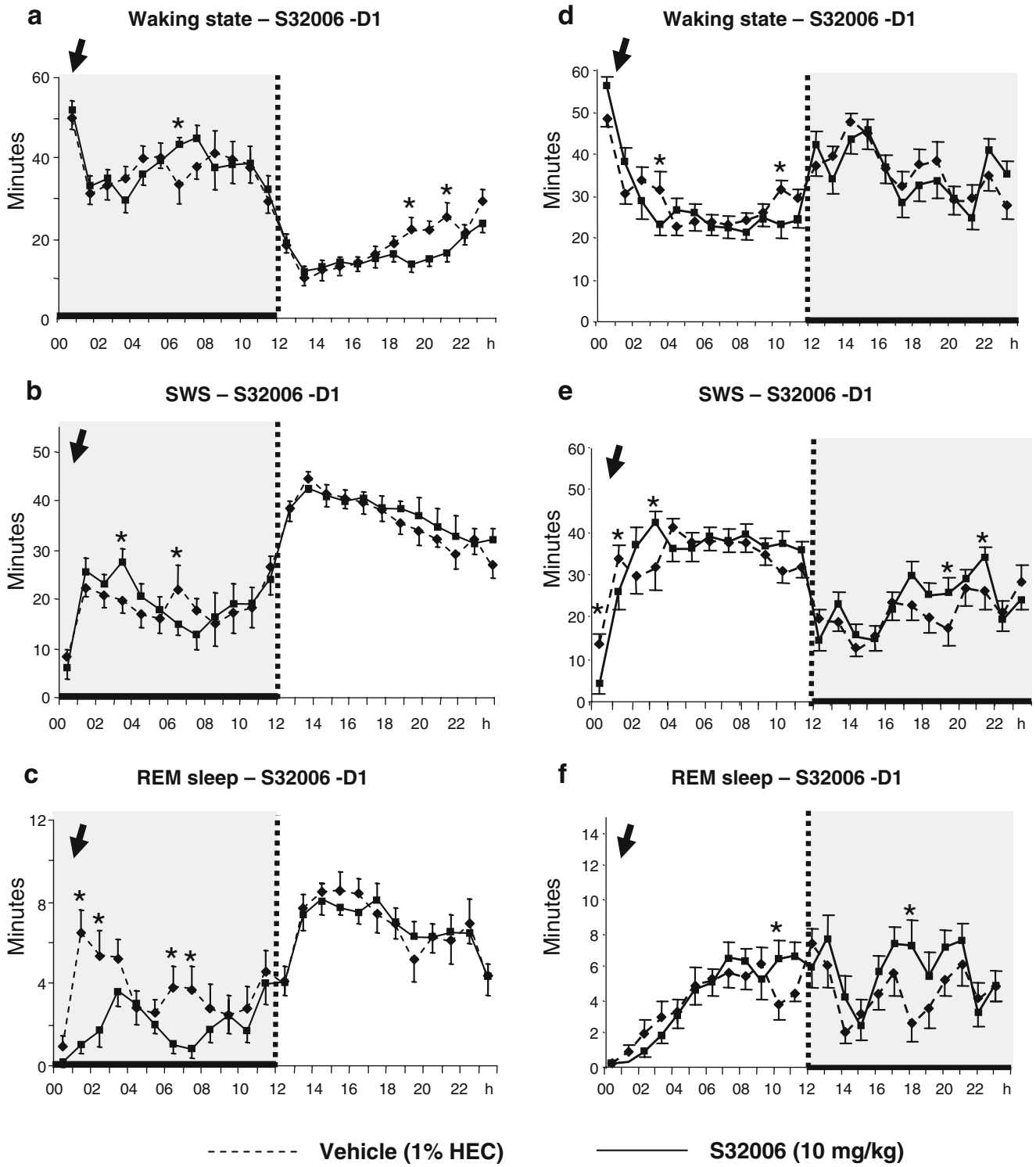


Fig. 4 Influence of S32006 (10.0 mg/kg, i.p.) upon 12 h measures of waking state (W, **a**,**d**), slow-wave sleep (SWS, **b**,**e**) and rapid eye movement sleep (REM, **c**,**f**) sleep following administration at the onset of the dark (**a**,**b**,**c**) or light (**d**,**e**,**f**) periods. S32006 was administered (*arrow*, 10.0 mg/kg, i.p.) at the onset of light (*right panel*), or dark (*left panel*) periods. Dark and light periods are indicated by *shaded* and *non-shaded parts* of the figure, respectively. Number of animals, 11 for vehicle (*1% HEC*) and S32006 when administered at the onset of the dark period, and 12 for vehicle (*1% HEC*) and S32006 when administered at the onset of the light period. *Ordinate* duration in minutes (each point represents mean duration per 1 h±SEM). *Abscissa* time-scale in hours (h), from "zero" which corresponds to the administration of S32006. *Asterisks* indicate significance (*$p<0.05$) of S32006 versus vehicle (1% HEC) differences in post-hoc least significant difference (LSD) tests following ANOVA. For other abbreviations, see also Fig. 1

circadian differences in the expression of 5-HT_{2C} receptor expression in various brain structures or a modulatory influence of other mechanisms: this remains to be elucidated (Holmes et al. 1997; Millan et al. 2005).

As mentioned above, the increased REM sleep seen with agomelatine cannot be attributed to 5-HT_{2C} blockade and it was marked at the lower of the two doses tested. Rather, this effect may be related to the melatoninergic properties of agomelatine at MT_1 and/or MT_2 receptors inasmuch as a similar action was seen for melatonin, even if transient and less pronounced. Furthermore, Cajochen et al. (1997) have reported that both melatonin and agomelatine increase REM sleep in healthy young men. This increase in REM sleep was most pronounced in the first REM sleep episode. One major structure expressing both MT_1 and MT_2 receptors is the suprachiasmatic nucleus that participates in the regulation of sleep–wake-function, at least partially via an indirect projection from to the noradrenergic locus coeruleus that controls arousal (Aston-Jones et al. 2001). However, the neuronal substrates underlying the influence of melatoninergic mechanisms upon REM sleep require direct evaluation. Mimicking the modest influence of melatonin upon REM sleep, a mild influence upon REM sleep (albeit limited in magnitude and duration as compared with agomelatine) was also seen with ramelteon, a synthetic and selective MT_1 and MT_2 receptor agonist used to treat insomnia. This effect, and its modest increase in SWS, corresponds to published data, though it is unclear why the influence of ramelteon and melatonin on SWS sleep and waking differ (Miyamoto et al. 2004; Erman et al. 2006; Borja and Daniel 2006).

The clear reduction in waking obtained with agomelatine was mimicked neither by melatonin nor by S32006 so it cannot be easily attributed to either its MT_1/MT_2 agonist or 5-HT_{2C} antagonist properties. Though underpinning the assertion that the impact of agomelatine upon sleep–wake architecture differs from both melatonin and the 5-HT_{2C} antagonist, S32006, the mechanistic basis of this decreased waking with agomelatine may be a compensatory response to increased REM and SWS sleep. A further possibility, though currently speculative, is that the influence of agomelatine upon waking and sleep–wake cycles in general should not be interpreted simply in terms of either melatonin "or" 5-HT_{2C} effects but rather their synergy, either functional and/or physical (Dugovic et al. 1989a, b; Millan 2005). This hypothesis is under exploration, but in this light, it is interesting that other actions of agomelatine such as its antidepressant properties in the learned helplessness test could be distinguished from both melatonin and 5-HT_{2C} antagonists alone (Bertaina-Anglade et al. 2006).

Possible clinical pertinence of findings: mood and cognition As pointed out in the "Introduction" section, many classes of antidepressant, including tricyclics, SSRIs, SNRIs, and MAO inhibitors, reduce REM sleep in patients (however, insomnia and the disruption of sleep architecture is observed only in 40–60% of outpatients with a diagnosis of major depression) and in rats but it remains controversial whether there is any causal relationship between this effect and improved mood (Winokur et al. 2001; Rijnbeek et al. 2003; Cespuglio et al. 2005; Millan 2006). Indeed, despite experimental and clinical evidence for antidepressant properties of agomelatine, it did not reduce REM sleep, mimicking the lack of REM suppression seen with mirtazapine (which shares the 5-HT_{2C} antagonist but not melatoninergic properties of agomelatine; Millan et al. 2000; Schittecatte et al. 2002). Inasmuch as agomelatine likewise did not diminish REM sleep in man (Quera Salva et al. 2007), these observations further suggest that reducing REM sleep may not be related to amelioration of the effect. Moreover, REM sleep may play a facilitatory role formation and maintenance of memory (Ishikawa et al. 2006; Fu et al. 2007), and cognitive processes are severely compromised in depressive patients yet little improved by standard drugs (Austin et al. 2001; Millan 2006). Accordingly, the augmenting influence of agomelatine upon REM sleep may tend to preserve cognitive performance. Though there is currently no evidence that agomelatine actually favors mnemonic function, this possibility would be of interest to examine. More generally, the present findings as well as other experimental and clinical evidence, suggest that agomelatine improves restorative SWS, sleep quality, and continuity. Agomelatine may, thus, be particularly useful in controlling the negative impact of persistent stress upon mood and circadian rhythmicity, possibility by blunting overactivity of the hypothalamo-pituitary-adrenal axis (Cespuglio et al. 1995; Marinesco et al. 1999; Lucassen et al. 2006; Millan 2006). This possibility is supported by studies showing that agomelatine moderates the disruption of diurnal rhythms of behavior, core temperature, and cortisol secretion associated with chronic social stress and counters the sleep fragmentation provoked by trypanosome infection in rats (Grassi-Zucconi et al. 1996; Corbach et al. 2007).

Conclusion This is the first quantitative EEG study of the influence of agomelatine upon sleep–wake architecture in rats. While agomelatine was ineffective when administered prior to the light period, when given before the dark period, agomelatine enhanced REM and SWS sleep. Inasmuch as melatonin tended to increase REM sleep, the melatonin agonist properties of agomelatine are likely related to the enhancement of REM sleep. By contrast, the promotion of SWS is probably due to 5-HT_{2C} receptor blockade inasmuch as S32006 mimicked this action upon administration prior to the dark (or light) phases. Intriguingly, the

reduction in waking seen with agomelatine differed from melatonin, ramelteon, and S32006, so it may reflect an interaction between actions at melatonin and 5-HT_{2C} receptors, though this remains to be clarified. Importantly, the influence of agomelatine upon sleep–wake architecture was expressed without a disruption of power spectra clearly differentiating it from hypnotics, and underpinning the contention that agomelatine is not a sleep-inducer agent per se, but rather affects circadian scheduling of sleep–wake cycles.

References

Armstrong SM, McNulty OM, Guardiola-Lemaître B, Redman JR (1993) Successful use of S20098 and melatonin in an animal model of delayed sleep-phase syndrome (DSPS). Pharmacol Biochem Behav 46:45–49

Aston-Jones G, Chen S, Zhu Y, Oshinsky ML (2001) A neural circuit for circadian regulation of arousal. Nat Neurosci 4:732–738

Audinot V, Mailliet F, Lahaye-Brasseur C, Bonnaud A, Le Gall A, Amosse C, Dromaint S, Rodriguez M, Nagel N, Galizzi JP, Malpaux B, Guillaumet G, Lesieur D, Lefoulon F, Renard P, Delagrange P, Boutin JA (2003) New selective ligands of human cloned melatonin MT_1 and MT_2 receptors. Naunyn-Schmiedeberg's Arch Pharmacol 367:553–561

Austin MP, Mitchell P, Goodwin GM (2001) Cognitive deficits in depression: possible implications for functional neuropathology. Br J Psychiatry 178:200–206

Baskett JJ, Broad JB, Wood PC, Duncan JR, Pledger MJ, English J, Arendt J (2003) Does melatonin improve sleep in older people? A randomized crossover trial. Age Ageing 32:164–170

Bertaina-Anglade V, La Rochelle CD, Boyer PA, Mocaer E (2006) Antidepressant-like effect of agomelatine in the learned helplessness model. Behav Pharmacol 17:703–713

Borja N, Daniel KL (2006) Ramelteon for the treatment of insomnia. Clin Therapeutics 28:1540–1555

Bourin M, Mocaër E, Porsolt R (2004) Antidepressant-like activity of S 20098 in the forced swimming test in rodents. Involvement of melatonin and 5-HT receptors. J Psy Neurosci 29:126–133

Cajochen C, Krauchi K, Graw P, Wirz-Justice A (1997) Melatonin and S-20098 increase REM sleep and wake-up propensity without modifying NREM sleep homeostasis. Am J Physiol 272:1189–1196

Cajochen C, Kräuchi K, Wirz-Justice A (2003) Role of melatonin in the regulation of human circadian rhythms and sleep. J Neuroendocrinol 15:432–437

Cespuglio R, Marinesco S, Baubet V, El Kafi B (1995) Evidence for a sleep-promoting influence of stress. Adv Neuroimmunol 5:145–154

Cespuglio R, Rousset C, Debilly G, Rochat C, Millan MJ (2005) Acute administration of the novel serotonin and noradrenaline reuptake inhibitor, S33005, markedly modifies sleep–wake cycle architecture in the rat. Psychopharmacology (Berl) 181:639–652

Chagraoui A, Protais P, Filloux T, Mocaër E (2003) Agomelatine (S20098) antagonizes the penile erections induced by the stimulation of 5-HT2C receptors in Wistar rats. Psychopharmacology (Berl) 170:17–22

Clement P, Gharib A, Cespuglio R, Sarda N (2003) Changes in sleep–wake cycle architecture and cortical NO release during aging in the rat. Neuroscience 116:863–870

Corbach S, Schmelting B, Fuchs E, Mocaër E (2007) Comparison of agomelatine and melatonin for effects in chronically stressed tree shrews, an animal model of depression. Eur Neurophyschopharmacol 17:S364

Dalton EJ, Rotondi D, Levitan RD, Kennedy SH, Brown GM (2000) Use of slow-release melatonin in treatment-resistant depression. J Psychiatry Neurosci 25:48–52

De Deurwaerdère P, Navaille S, Berg KA, Clarke WP, Spampinato U (2004) Constitutive activity of the serotonin2C receptor inhibits in vivo dopamine release in the rat striatum and nucleus accumbens. J Neurosci 24:3235–3241

Dekeyne A, Mannoury La Cour C, Gobert A, Brocco M, Lejeune F, Serres F, Sharp T, Daszuta A, Soumier A, Papp M, Rivet JM, Flik G, Cremers TI, Muller O, Lavielle G, Millan MJ (2008) S32006, a novel 5-HT_{2C} receptor antagonist displaying broad-based antidepressant and anxiolytic properties in rodent models. Psychopharmacology (Berl) 199(4):549–568

Dugovic C, Leysen JE, Wauquier A (1989a) Melatonin modulates the sensitivity of 5-hydroxytryptamine-2 receptor-mediated sleep–wakefulness regulation in the rat. Neurosci Lett 104:320–325

Dugovic C, Wauquier A, Leysen JE, Marrannes R, Jannsen PA (1989b) Functional role of 5-HT2 receptors in the regulation of sleep and wakefulness in the rat. Psychopharmacology (Berl) 97:436–442

Erman M, Seiden D, Zammit G, Sainati S, Zhang J (2006) An efficacy, safety and dose-response study of ramelteon in patients with chronic primary insomnia. Sleep 7:17–24

Fu CH, Williams SC, Brammer MJ, Suckling J, Kim J, Cleare AJ, Walsh ND, Mitterschiffthaler MT, Andrew CM, Pich EM, Bullmore ET (2007) Neural responses to happy facial expressions in major depression following antidepressant treatment. Am J Psychiatry 164:599–607

Gibbs FP, Uriend J (1981) The half life of melatonin elimination from rat plasma. Endocrinology 109:1796–1798

Gorka Z, Moryl E, Papp M (1996) Effect of chronic mild stress on circadian rhythms in the locomotor activity in rats. Pharmacol Biochem Behav 54:229–234

Grassi-Zucconi G, Semprevivo M, Mocaer E, Kristensson K, Bentivoglio M (1996) Melatonin and its new antagonist S-20098 restore synchronized sleep fragmented by experimental trypanosome infection in the rat. Brain Res Bull 39:63–68

Hack LM, Lockley SW, Arendt J, Skene DJ (2003) The effects of low-dose 0.5-mg melatonin on the free-running circadian rhythms of blind subjects. J Biol Rhythms 18:420–429

Holmes MC, French KL, Seckl JR (1997) Dysregulation of diurnal rhythms of serotonin 5-HT2C and corticosteroid gene expression in the hippocampus with food restriction and glucocorticoids. J Neurosci 17:4056–4065

Huber R, Deboer T, Schwierin B, Tobler I (1998) Effect of melatonin on sleep and brain temperature in the Djungarian hamster and the rat. Physiol Behav 65:77–82

Hughes RJ, Sack RL, Lewy AJ (1998) The role of melatonin and circadian phase in age-related sleep-maintenance insomnia: assessment in a clinical trial of melatonin replacement. Sleep 21:52–68

Ishikawa A, Kanayama Y, Matsumura H, Tsuchimochi H, Ishida Y, Nakamura S (2006) Selective rapid eye movement sleep deprivation impairs the maintenance of long-term potentiation in the rat hippocampus. Eur J Neurosci 24:243–248

Kantor S, Jakus R, Bodizs R, Halasz P, Bagdy G (2002) Acute and long-term effects of the 5-HT2 receptor antagonist ritanserin on EEG power spectra, motor activity, and sleep: changes at the light-dark phase shift. Brain Res 943:105–111

Karim A, Tolbert D, Cao C (2006) Disposition kinetics and tolerance of escalating single doses of ramelteon, a high-affinity MT1 and

MT2 melatonin receptor agonist indicated for treatment of insomnia. J Clin Pharmacol 46(2):140–148
Kato K, Hirai K, Nishiyama K, Uchikawa O, Fukatsu K, Ohkawa S, Kawamata Y, Hinuma S, Miyamoto M (2005) Neurochemical properties of ramelteon (TAK-375), a selective MT1/MT2 receptor agonist. Neuropharmacology 48:301–310
Kennedy SH, Emsley R (2006) Placebo-controlled trial of agomelatine in the treatment of major depressive disorder. Eur Neuropsychopharmacol 16:93–100
Lemoine P, Guilleminault C, Alvarez E (2007) Improvement in subjective sleep in major depressive disorder with a novel antidepressant, agomelatine: randomized, double-blind comparison with venlafaxine. J Clin Psychiatry 68:1723–1732
Leproult R, Van Onderbergen A, L'Hermite-Baleriaux M, Van Cauter E, Copinschi G (2005) Phase-shifts of 24-h rhythms of hormonal release and body temperature following early evening administration of the melatonin agonist agomelatine in healthy older men. Clin Endocrinol (Oxf) 63:298–304
Lewy AJ, Ahmed S, Jackson JM, Sack RL (1992) Melatonin shifts human circadian rhythms according to a phase-response curve. Chronobiol Int 9:380–392
Lockley SW, Skene DJ, James K, Thapan K, Wright J, Arendt J (2000) Melatonin administration can entrain the free-running circadian system in blind subjects. J Endocrinol 164:R1–R6
Lucassen PJ, Heine VM, Muller MB, van der Beek EM, Wiegant VM, De Kloet ER, Joels M, Fuchs E, Swaab DF, Czeh B (2006) Stress, depression and hippocampal apoptosis. CNS Neurol Disord Drug Targets 5:531–546
Mahlberg R, Kunz D (2007) Melatonin excretion levels and polysomnographic sleep parameters in healthy subjects and patients with sleep-related disturbances. Sleep Med 8:512–516
Marinesco S, Bonnet C, Cespuglio R (1999) Influence of stress duration on the sleep rebound induced by immobilization in the rat: a possible role for corticosterone. Neuroscience 92:921–933
Martinet L, Guardiola-Lemaître B, Mocaër E (1996) Rhythms by S 20098, a melatonin agonist, is dose and plasma concentration dependent. Pharmacol Biochem Behav 54:713–718
Millan MJ (2005) Serotonin 5-HT2C receptors as a target for the treatment of depressive and anxious states: focus on novel therapeutic strategies. Therapie 60:441–460
Millan MJ (2006) Multi-target strategies for the improved treatment of depressive states: Conceptual foundations and neuronal substrates, drug discovery and therapeutic application. Pharmacol Ther 110:135–370
Millan MJ, Lejeune F, Gobert A (2000) Reciprocal autoreceptor and heteroreceptor control of serotoninergic, dopaminergic and noradrenergic transmission in the frontal cortex: relevance to the actions of antidepressant agents. J Psychopharmacol 14:114–138
Millan MJ, Gobert A, Lejeune F, Dekeyne A, Newman-Tancredi A, Pasteau V, Rivet JM, Cussac D (2003) The novel melatonin agonist agomelatine (S20098) is an antagonist at 5-hydroxytryptamine2C receptors, blockade of which enhances the activity of frontocortical dopaminergic and adrenergic pathways. J Pharmacol Exp Ther 306:954–64
Millan MJ, Brocco M, Gobert A, Dekeyne A (2005) Anxiolytic properties of agomelatine, an antidepressant with melatoninergic and serotonergic properties: role of 5-HT2c receptors blockade. Psychopharmacology (Berl) 177:448–458
Miyamoto M, Nishikawa H, Doken Y, Hirai K, Uchikawa O, Ohkawa S (2004) The sleep-promoting action of ramelteon (TAK-375) in freely moving cats. Sleep 27(7):1319–1325
Monti JM, Jantos H (2006) Effects of the serotonin 5-HT2A/2C receptor agonist DOI and of the selective 5-HT2A or 5-HT2C receptor antagonists EMD 281014 and SB-243213, respectively, on sleep and waking in the rat. Eur J Pharmacol 553:163–170
Mundey K, Benloucif S, Harsanyi K, Dubocovich ML, Zee PC (2005) Phase-dependent treatment of delayed sleep phase syndrome with melatonin. Sleep 28(10):1214–1216
Olié JP, Kasper S (2007) Efficacy of agomelatine, a MT1/MT2 receptor agonist with 5-HT2C antagonistic properties, in major depressive disorder. Int J Neuropsychopharmacol 10:661–673
Pandi-Perumal SR, Srinivasan V, Poeggeler B, Hardeland R, Cardinali DP (2007) Drug insight: the use of melatoninergic agonists for the treatment of insomnia-focus on ramelteon. Nat Clin Pract Neurol 3:221–228
Papp M, Gruca P, Boyer P-A, Mocaer E (2003) Effect of agomelatine in the chronic mild stress model of depression in the rat. Neuropsychopharmacology 28:694–703
Paxinos G, Watson C (1998) The rat brain in stereotaxic coordinates. Academic, San Diego
Quera Salva MA, Vanier B, Laredo J, Hartley S, Chapotot F, Moulin C, Lofaso F, Guilleminault C (2007) Major depressive disorder, sleep EEG and agomelatine: an open-label study. Int J Neuropsychopharmacol 10:691–696
Rajaratnam SM, Dijk DJ, Middleton B, Stone BM, Arendt J (2003) Melatonin phase-shifts human circadian rhythms with no evidence of changes in the duration or endogenous melatonin secretion or the 24-hour production of reproductive hormones. J Clin Endocrinol Metab 88:4303–4309
Redman JR, Francis AJP (1998) Entrainment of rat circadian rhythms by the melatonin agonist S 20098 requires intact suprachiasmatic nuclei but not the pineal. J Biological Rhythms 13:39–51
Redman JR, Guardiola-Lemaître B, Brown M, Delagrange P, Armstrong SM (1995) Dose-dependent effects of S 20098, a melatonin agonist, on direction of re-entrainment of rat circadian activity rhythms. Psychopharmacology (Berl) 118:385–390
Rijnbeek B, de Visser SJ, Franson KL, Cohen AF, van Gerven JM (2003) REM sleep effects as a biomarker for the effects of antidepressants in healthy volunteers. J Psychopharmacol 17:196–203
Roth T, Stubbs C, Walsh JK (2005) Ramelteon (TAK-375), a selective MT1/MT2-receptor agonist, reduces latency to persistent sleep in a model of transient insomnia related to a novel sleep environment. Sleep 28:303–307
Schittecatte M, Dumont F, Machowski R, Cornil C, Lavergne F, Wilmotte J (2002) Effects of mirtazapine on sleep polygraphic variables in major depression. Neuropsychobiology 46:197–201
Sharpley AI, Cowen PJ (1995) Effect of pharmacologic treatments on the sleep of depressed patients. Biol Psychiatry 37:85–98
Sharpley AL, Elliott JM, Attenburrw MJ, Cowen PJ (1994) Slow wave sleep in humans: role of 5-HT2A and 5-HT2C receptors. Neuropharmacology 33:467–471
Slotten HA, Krekling S, Sicard B, Pévet P (2002) Daily infusion of melatonin entrains circadian activity rhythms in the diurnal rodent Arvicanthis ansorgei. Behav Brain Res 133:11–19
Smith MI, Piper DC, Duxon MS, Upton N (2002) Effect of SB-243213, a selective $5HT_{2c}$ receptor antagonist on the rat sleep profile: a comparison to paroxetine. Pharmacol Biochem Behav 71:599–605
Tobler I, Jaggi K, Borbely AA (1994) Effects of melatonin and the melatonin receptor agonist S-20098 on the vigilance states, EEG spectra and cortical temperature in the rat. J Pineal Res 16:26–32
Trivedi MH, Rush AJ, Armitage R, Gullion CM, Grannemann BD, Orsulak PJ, Roffwarg HP (1999) Effects of fluoxetine on polysomnogram in outpatients with major depression. Neuropsychopharmacology 20:447–59
Ursin R (2002) Serotonin and sleep. Sleep Med Rev 6:57–69
Van Reeth O, Olivares E, Zhang Y, Zee PC, Mocaer E, Defrance R, Turek FW (1997) Comparative effects of a melatonin agonist on the circadian system in mice and Syrian hamsters. Brain Res 762 (1–2):185–194

Vogel GW, Buffenstein A, Minter K, Hennessey A (1990) Drug effects on REM sleep and on endogenous depression. Neurosci Biobehav Rev 14:49–63

Wade AG, Ford I, Crawford G, McMahon AD, Nir T, Laudon M, Zisapel N (2007) Efficacy of prolonged release melatonin in insomnia patients aged 55–80 years: quality of sleep and next-day alertness outcomes. Curr Med Res Opin 23:2597–2605

Winokur A, Gary KA, Rodner S, Rae-Red C, Fernando AT, Szuba MP (2001) Depression, sleep physiology, and antidepressant drugs. Depress Anxiety 14:19–28

Wirz-Justice A, Van den Hoofdakker RH (1999) Sleep deprivation in depression: what do we know, where do we go? Biol Psychiatry 15:445–453

Zemlan FP, Mulchahey JJ, Scharf MB, Mayleben DW, Rosenberg R, Lankford A (2005) The efficacy and safety of the melatonin agonist beta-methyl-6-chloromelatonin in primary insomnia: a randomized, placebo-controlled, crossover clinical trial. J Clin Psychiatry 66:384–390

Zupancic M, Guilleminault C (2006) Agomelatine: a preliminary review of a new antidepressant. CNS Drugs 20:981–992