Psychopharmacology (1998) 140:503–509

ORIGINAL INVESTIGATION

Jenny L. Wiley · Mario E. Dance · Robert L. Balster

Preclinical evaluation of the reinforcing and discriminative stimulus effects of agomelatine (S-20098), a melatonin agonist

Received: 26 March 1998 / Final version: 27 May 1998

Abstract Agomelatine (S-20098), an analog of melatonin, has shown promise as a chronobiotic in animal models of sleep phase disorders and is being developed for clinical use. Previous research has shown that the pharmacological profile of melatonin-like drugs overlaps that of γ-amino butyric acid (GABA) agonists. Given the potential of drugs within the latter class for recreational abuse in humans, evaluation of this potential for melatonin analogs that target similar therapeutic indications is important. In the present study, agomelatine was tested in animal models of the subjective and reinforcing effects of CNS depressant drugs; i.e., diazepam discrimination in rats and IV methohexital self-administration in rhesus monkeys, respectively. Neither agomelatine nor melatonin substituted for diazepam in rats trained to discriminate 2.5 mg/kg diazepam from vehicle. Further, agomelatine was not self-administered by rhesus monkeys. These results suggest that agomelatine would not produce diazepam-like intoxication in humans, nor would it likely be subject to abuse.

Key words Agomelatine · S-20098 · Melatonin · Diazepam · Methohexital · Drug discrimination · Self-administration · Substance abuse evaluation · Monkey · Rat

Introduction

Melatonin, an endogenous substance secreted by the pineal gland, has a number of purported physiological effects. Under normal circumstances, it is produced in humans and other mammals primarily at night. In animals, melatonin regulates circadian and diurnal rhythms, is involved in seasonal reproduction patterns, and may have hypnotic effects (Golombek et al. 1996; Reppert et al. 1996). Its exact physiological roles in humans are unknown. At least two sub-types of melatonin receptors have been identified and, in most mammals, these receptors are localized primarily in the suprachiasmatic nucleus (SCN; the site of the biological clock) and in the retina. When administered IV to humans, melatonin is lipophilic and is rapidly distributed to the brain, but has a short half-life of 15–30 min (Claustrat et al. 1989). Although melatonin is under investigation for a number of therapeutic implications, including treatment of sleep phase disorders, "jet lag", and seasonal affective disorder (Attenburrow et al. 1995; Hagan and Oakley 1995; Brzezinski 1997), its non-selective binding at its different target sites and its short half-life make it less than an ideal chronobiotic candidate. To address these pharmacokinetic problems of melatonin, several metabolically more stable analogs have been synthesized (Depreux et al. 1994).

One of these melatonin analogs, agomelatine, *N*[2-(>-methoxy-naphet-I-yl)ethyl]acetamid, a naphtalenic bioisoster of melatonin, has been selected for clinical development (Depreux et al. 1994). Agomelatine binds to ovine pars tuberalis melatonin receptors with affinity similar to that of melatonin (K_D=1.00±0.35 and 9.15± 3.98, respectively; Depreux et al. 1994). Previous studies in rats have shown that agomelatine shares pharmacological effects with melatonin, including dose-dependent entrainment of circadian rhythms and phase advancement of activity onset in an animal model of delayed sleep-phase syndrome (Armstrong et al. 1993; Tobler et al. 1994; Redman et al. 1995; Martinet et al. 1996). Further, agomelatine was approximately equipotent to melatonin in producing these effects. The range of agomelatine doses tested in the present study (1–100 mg/kg, IP) was inclusive of those that were active in these phase-shift procedures (1–10 mg/kg, oral or IP). Hence, preclinical evaluation of this drug suggests that, similar to melatonin, agomelatine may have therapeutic efficacy as a chronobiotic in the treatment of sleep disorders and other types of conditions that may involve disruption of biological rhythms.

Previous research has provided evidence for an interaction between melatonin and GABAergic systems. Melatonin shares several pharmacological effects in rodents

J.L. Wiley (✉) · M.E. Dance · R.L. Balster
Virginia Commonwealth University, Medical College of Virginia,
Department of Pharmacology and Toxicology,
Richmond, VA 23298-0613, USA

with benzodiazepines and other $GABA_A$ agonists, including sedative, anxiolytic, and anticonvulsant effects (Golombek et al. 1993, 1996; Pierrefiche et al. 1993). In addition, Levesque and Locke (1994) found that the benzodiazepine, triazolam, fully substituted for melatonin in rats trained to discriminate melatonin (150 mg/kg, IP) from saline in a two-lever drug discrimination procedure. In addition, because therapeutic development of melatonin agonists may target indications such as sedation, where existing medications can develop problems with abuse, it is important to obtain information on their abuse potential. The purpose of the present study was two-fold: (1) to investigate further similarities and differences in the discriminative stimulus effects of melatonin-like drugs and the benzodiazepine diazepam in rats and (2) to evaluate agomelatine for abuse potential in an IV methohexital self-administration procedure in rhesus monkeys. This latter procedure is an animal model of the reinforcing effects of drugs in humans, with a generally good correlation between those drugs that are self-administered by laboratory animals and those that are recreationally abused by humans (Johanson and Balster 1978; Ator and Griffiths 1987).

Materials and methods

Rat drug discrimination

Ten adult male Sprague-Dawley rats, obtained from Harlan (Dublin, Va., USA), were food-restricted and allowed to gain weight slowly over the course of the experiment. Rats were maintained at body weights between 350 and 400 g by rationing the daily amount of Lab Diet #5000 rodent chow (PMI Nutrition International, St Louis, Mo., USA) received after experimental sessions. When sessions were not being conducted, the rats were individually housed in wire suspension cages in a temperature-controlled (20–22°C) vivarium environment with a 12-h light-dark cycle (lights on at 7 a.m.). Water was freely available in the home cages. Rats were drug naive and were approximately 3 months old at the beginning of the experiment. They were weighed daily and acclimated to the laboratory environment for 1 week prior to the beginning of drug discrimination training.

During behavioral training and testing, rats were transported from the vivarium to a laboratory in the same building, injected and placed into standard two-lever operant conditioning chambers (Lafayette Instruments Co., Lafayette, Ind., USA) equipped with stimulus lights and a dispenser capable of delivering 45 mg Bio-Serv (Frenchtown, N.J., USA) food pellets. They were trained to discriminate 2.5 mg/kg diazepam from an equal volume of vehicle (1:4:5 mixture of ethanol, propylene glycol, and distilled water). A standard two-lever drug discrimination procedure, as used previously (Wiley et al. 1995), was employed. Rats were trained during daily (Monday to Friday) sessions, of 15 min duration. Sessions were conducted between 1400 and 1600 hours during the light portion of the light-dark cycle. After rats had learned to press the levers in order to obtain food reinforcement, they were injected with either diazepam (2.5 mg/kg) or its vehicle in a double alternation schedule. On days when diazepam was administered, lever presses on the drug-associated lever were reinforced under a fixed-ratio (FR) 10 schedule. On days when vehicle was administered, lever presses on the other (vehicle-associated) lever were reinforced. In either case, the rat was required to make ten consecutive responses on the injection-appropriate lever in order to receive food reinforcement. Responses on the incorrect lever reset the ratio requirement on the correct lever.

After rats had acquired the discrimination, test sessions were conducted on Tuesdays and Fridays if the subject completed the first fixed-ratio on the correct lever during the preceding training session. During test sessions, ten consecutive responses on either lever were reinforced. Between test sessions, the rats continued the double alternation sequence of diazepam and vehicle training sessions. Rats were injected with one of the test compounds (diazepam, agomelatine or melatonin) or with vehicle 30 min before the start of a test session. The dose-effect curve for diazepam was determined first, followed by the dose-effect curve determination for agomelatine. Melatonin was tested last. Doses of each drug were tested in ascending order. Before the start of each dose-effect curve, rats were administered control tests with vehicle and with 2.5 mg/kg diazepam. In addition, a test with 5 ml/kg volume of the vehicle for agomelatine was tested following completion of the dose-effect curve determination for this drug.

Rhesus monkey self-administration

Four adult male rhesus monkeys (*Macaca mulatta*), weighing 7.8–12.1 kg, were housed in 1-m^3 fiberglass cubicles with a transparent front door. The monkeys were fed Purina Monkey Chow twice daily (morning and evening) and received ad libitum water in their home cages. Dietary supplements consisting of a chewable multiple vitamin tablet, Prima Treats (BioServ), and fresh fruits or vegetables were provided daily. All monkeys had previously participated in other self-administration studies.

Each monkey had previously been surgically implanted with indwelling silicone catheters (0.08 i.d., Ronsil Rubber Products, Belle Mead, N.J., USA) under phencyclidine (1 mg/kg, IM)/ pentobarbital (10–30 mg/kg, IV) anesthesia. Catheters ran SC from the catheterized vein and exited in the mid-scapular area. They were protected by stainless steel harnesses and restraining arms through which the catheters passed to the rear of the cubicles. The catheter-protection harness and tether were equipped with swivels allowing animals nearly complete freedom of movement within the cubicles. A peristaltic pump (Masterflex, Cole-Palmer, Chicago, Ill., USA) was attached to each catheter and delivered 1-ml infusions in 10 s. Two response levers and associated stimulus lights were located on the front door of each cubicle. Experimental contingencies and data recording were accomplished by a PCP-11 computer located in an adjacent room and utilizing SKED-11 software (State Systems, Kalamazoo, Mich., USA).

Each monkey was trained to press the left lever for 0.1 mg/kg per injection sodium methohexital under a fixed ratio 10 (FR-10) schedule of reinforcement during daily 1-h experimental sessions. Daily sessions began at approximately 1400 hours during the light portion of the light-dark cycle. Availability of the drug was signaled by illumination of two white stimulus lights above the lever. During infusions, the white lights were extinguished and a red light located between them was illuminated. When rates of methohexital self-administration were stable, substitution tests with methohexital and agomelatine were performed. Tests were conducted with saline before and after the testing of agomelatine and a test of the agomelatine vehicle was conducted as part of the agomelatine dose-effect curve determination. Substitution tests comprised four consecutive sessions in which a test solution was made available for self-administration. No external stimuli were presented to the monkey to indicate that the solution had been changed. Between substitution tests, monkeys were returned to methohexital self-administration for a minimum of 3 days until stable performance was again obtained.

Drugs

For the rat discrimination study, diazepam (Elkin-Sinn, Cherry Hill, N.J., USA) was purchased commercially in a concentration of 5 mg/ml. A vehicle of ethanol: propylene glycol: distilled water in a 1:4:5 volume ratio was used to dilute this stock concentration to lower doses. Doses up to 5 mg/kg were administered in a vol-

ume of 1 ml/kg. Higher doses were obtained by adjusting the injection volume. Agomelatine (formerly known as S-20098; Institut de Recherches Internationale Servier, Courbevoie, France) and melatonin (Research Biochemicals International, Natick, Mass., USA) were dissolved in 1:4:5 ratio of ethanol:polyethylene glycol:distilled water. Each drug was injected at a volume of 1 ml/kg, with the exception that the 100 mg/kg dose of agomelatine was obtained by injecting 5 ml/kg of a 20 mg/ml concentration. Each of the three test drugs was injected IP 30 min before the start of the session.

For the monkey study, sodium methohexital was obtained commercially as Brevital (500 mg/10 ml vial; Jones Medical Industries, Inc., St Louis, Mo., USA). The stock solution was diluted with 0.9% sterile saline to produce test concentrations. Agomelatine was dissolved in pure ethanol to produce a concentration of 100 mg/ml. This ethanolic solution was then mixed in a 1:1 ratio with a polyoxyethylated vegetable oil (emulphor; EL-620; Rhone Poulenc, Princeton, N.J., USA). Micellar suspensions were then formed by mixing the ethanol:emulphor solution with 0.9% saline to produce desired test concentrations and were delivered in a volume of 1 ml/kg per infusion. For vehicle test solutions, the highest concentration of ethanol:emulphor:saline needed to solubilize the 1 mg/kg per infusion test dose of agomelatine was used.

Data analysis

For the rat study, mean (±SEM) percentage of responses on the diazepam-associated lever and mean (±SEM) overall rate of responding (responses/s) were calculated for each test session separately. When appropriate, ED_{50}s (with 95% confidence intervals) were calculated for each drug using least-squares linear regression on the linear part of the dose-effect curves (Tallarida and Murray 1987) for percentage of drug-lever responding, plotted against $\log_{10}$ transformation of the dose. Data on percentage of diazepam-lever responding for rats that responded less than 0.02 responses/s at a particular drug dose were not included in the mean calculation for this measure. A minimum of 0.02 responses/s was necessary in order for a rat to receive a reinforcer during a test session (i.e., complete the fixed ratio requirement and choose a lever). Response rate data for all rats were included in the calculation of mean response rate. Separate repeated measures ANOVAs (with Tukey post hoc tests at α=0.05) were used to evaluate significant changes in response rates (compared to vehicle control rates).

For each monkey, mean (±range) injection rate during the last 3 days of each 4-day agomelatine or methohexital dose substitution was calculated. Methohexital control data consisted of the mean (±SD) number of methohexital infusions during the three baseline sessions which preceded each substitution test (total=36). Vehicle control data was calculated as the mean (±range) on the last 3 days before each substitution test. Data are presented separately for each monkey.

Results

Discriminative stimulus effects in rats

As expected, diazepam produced full dose-dependent substitution for the training dose (Fig. 1a), indicating that rats had successfully acquired the discrimination. The ED_{50} for diazepam substitution was 0.84 mg/kg (95% CL: 0.68–1.06 mg/kg). Significant response rate suppression occurred at the 15 mg/kg dose of diazepam (F6,48=9.1, P<0.0001) (Fig. 1b). Melatonin and agomelatine failed to substitute for diazepam at any dose (Fig. 1a). Response rates were significantly reduced at the 100 mg/kg dose of each drug (melatonin: F3,24=9.5, P=0.004; agomelatine: F5,45=5.2, P=0.001) (Fig. 1b), indicating that behaviorally active doses were tested. Throughout the study, responding during control tests with diazepam (DZP) and the vehicle for diazepam (VEH) occurred almost exclusively on the injection-appropriate lever, as did responding during a control test with a 5 ml/kg volume of the vehicle for agomelatine (5XV) (as used for the highest dose of agomelatine).

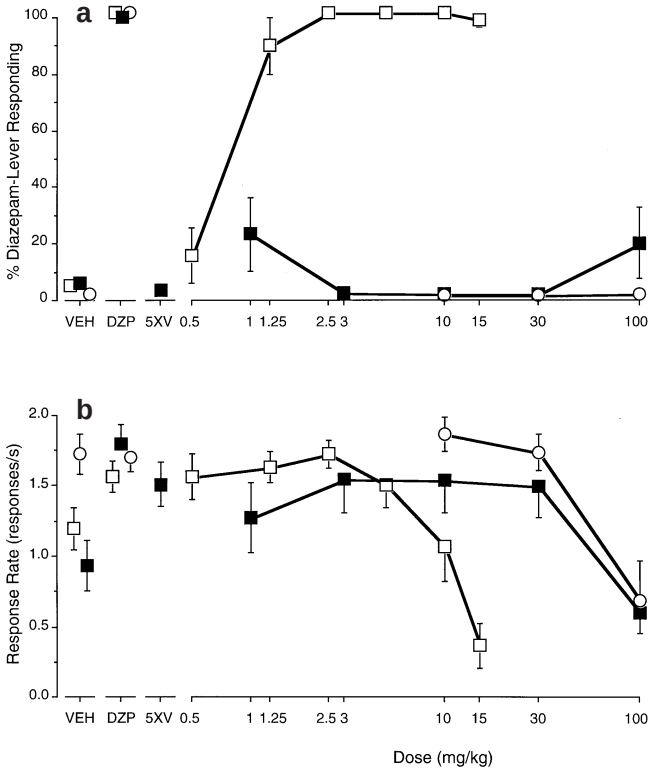

Fig. 1a, b Effects of diazepam (□), agomelatine (■) and melatonin (○) on percentage of diazepam-lever responding (**a**) and response rates (**b**) in rats trained to discriminate diazepam (2.5 mg/kg) from vehicle. Points above *VEH* and *DZP* represent the results of control tests with vehicle and 2.5 mg/kg diazepam conducted before each dose-effect curve determination. Points above *5XV* represent the results of a control test with 5 ml/kg volume of vehicle. Each value represents the mean (±SEM) of nine to ten rats, except for percentage of diazepam-lever responding for the 15 mg/kg dose of diazepam (n=5), the 100 mg/kg dose of agomelatine (n=8) and the 100 mg/kg dose of melatonin (n=6)

Self-administration in rhesus monkeys

Figure 2 presents the results of substitution tests with methohexital and agomelatine in rhesus monkeys trained to self-administer 0.1 mg/kg methohexital. Under baseline conditions, 0.1 mg/kg methohexital (MET) was readily self-administered by each monkey with average injection rates of 40–80/session. When saline (S1 and S2) was substituted for methohexital, injection rates decreased substantially, although there was some variability in saline substitution rates, especially in monkey M1145. Results of the dose-effect curve determination with metho-

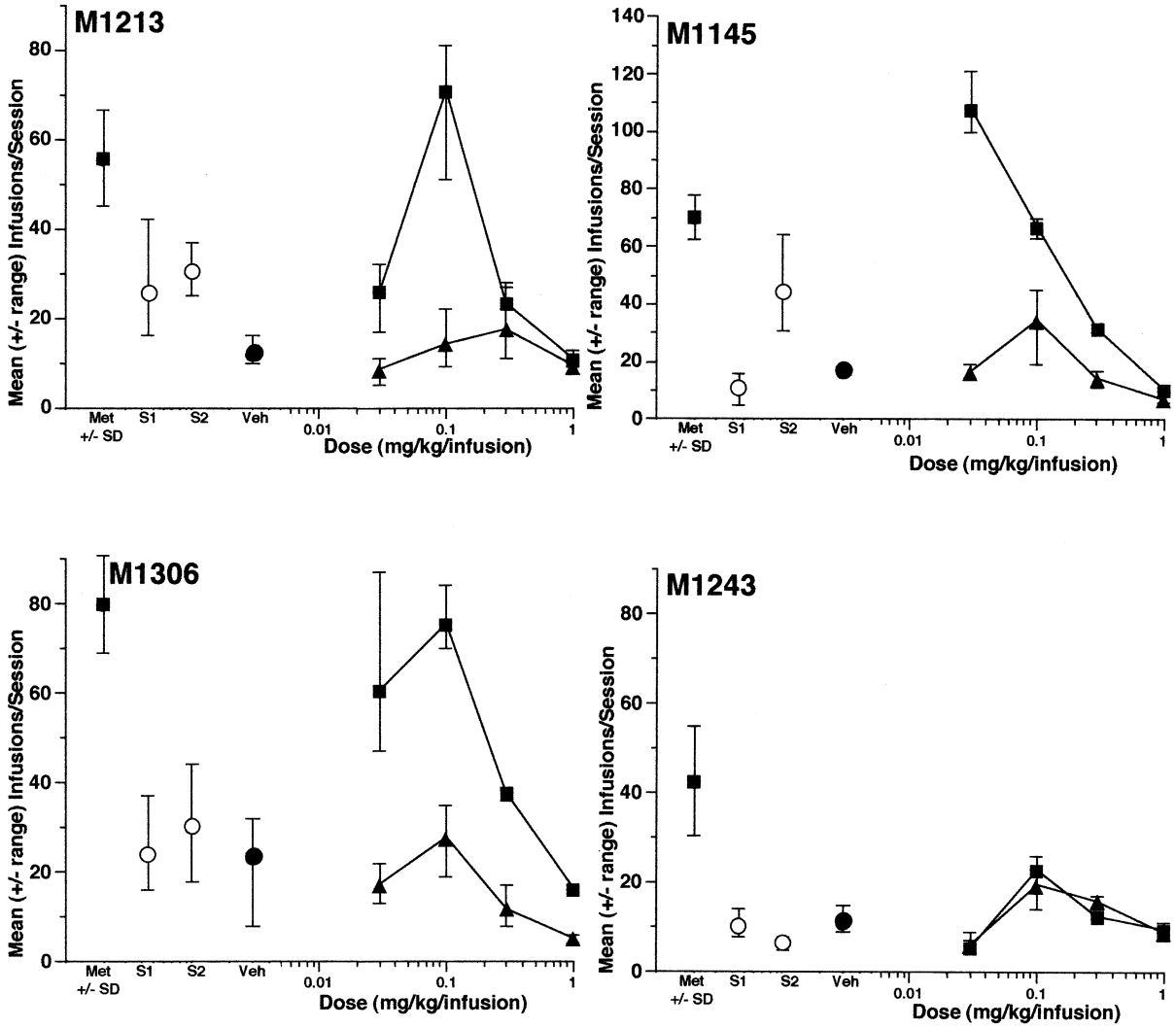

Fig. 2 Mean (±range) number of infusions of methohexital and agomelatine in rhesus monkeys trained to self-administer IV methohexital (0.1 mg/kg per infusion). Points above *S1* and *S2* (○) represent the results of control tests with saline conducted before and after the dose-effect curve determination for agomelatine. Points above *VEH* (●) represent the results of substitution tests with the vehicle for agomelatine. Points above *MET* (■) represent the mean (±SD) for the 36 baseline methohexital tests conducted throughout the study. Data are shown individually for each monkey

hexital revealed that, for each monkey, infusion rates for at least one dose of methohexital exceeded rates during all saline control tests and the ranges did not overlap, presenting clear evidence for reinforcing effects of this drug. For monkeys M1306, M1213 and M1243, there was an inverted U-shaped dose-effect curve relating dose per infusion to infusion rate. It is likely that a similar dose-effect curve would have been obtained for monkey M1145, were a lower dose tested.

In contrast to the results with methohexital, there was no evidence for agomelatine self-administration. For monkeys M1306, M1213 and M1145, rates of agomelatine self-administration fell well within the rates observed under saline and vehicle substitution test conditions. Although the mean infusion rate for the 0.1 mg/kg dose of agomelatine slightly exceeded the mean rates for saline and vehicle in monkey M1243, the range for the vehicle and S1 saline tests were overlapping with the range for this test dose of agomelatine. For monkey M1145, the infusion rate for the 0.1 mg/kg dose of agomelatine was also higher than that sometimes obtained for saline and vehicle; however, this monkey showed occasional saline substitution rates with equally high rates (e.g., see S2 rates). At the highest test dose of agomelatine, there are some instances where injection rates were suppressed below those for saline and vehicle. This is most clear for M1306, where the injection rates at 1 mg/kg are below the ranges for both saline and the vehicle tests. In M1145, injection rates for this dose of agomelatine are below those for two of the three control tests.

Discussion

In the drug discrimination study, both agomelatine and melatonin completely failed to substitute for diazepam.

Response-rate decreasing effects were obtained, showing that a behaviorally active dosage range was tested. These results with the melatonin agonists are in contrast with results typically obtained in substitution tests with abused CNS depressant drugs in benzodiazepine-trained animals (Ator and Griffiths 1987). In general, all benzodiazepine agonists cross-substitute for one another with potencies predicted by their affinity for benzodiazepine receptors (Young and Glennon 1987). In addition, cross substitution is usually found among benzodiazepines and barbiturates (Ator and Griffiths 1997), reflective of the similarities in the acute intoxications produced by these classes of drugs (de Wit and Griffiths 1991). Because of this, our results would allow us to predict that melatonin and agomelatine would not produce acute subjective effects similar to those of the benzodiazepines or barbiturates and to conclude that they would not have abuse potential of the CNS depressant type. This is the general rationale used for including drug discrimination tests in preclinical abuse potential evaluation (Balster 1990; Holtzman 1990).

On the other hand, our data showing no diazepam-like discriminative stimulus effects with agomelatine and melatonin are not consistent with results obtained by Levesque and Locke (1994) in which rats trained to discriminate melatonin from saline showed full substitution with triazolam, flurazepam and pentobarbital. Asymmetrical cross-generalization is sometimes obtained in drug discrimination studies, and has even been reported for benzodiazepines and barbiturates, where barbiturates can fail to substitute in lorazepam-trained animals (Ator and Griffiths 1989). But among barbiturates and benzodiazepines, this is the unusual case (Ator and Griffiths 1997). One possible explanation for the asymmetrical cross-generalization results with melatonin resides in the doses used in the two studies. The highest dose of melatonin we tested was 100 mg/kg, which produced substantial decreases in rates of responding. Levesque and Locke (1994) trained their rats with 150 mg/kg melatonin, a dose that they reported produced sedative and muscle relaxant effects. It is likely that they were able to administer this high dose of melatonin without severe disruption of responding because they utilized a shock-avoidance procedure which is often more resistant to drug disruption than the food-reinforced responding used in our study. Unfortunately, response rates are not reported to allow a determination to be made if the cross-substitution occurred at doses that produced substantial direct behavioral effects which may have disrupted the discrimination. In any case, it is clear from our study that melatonin and agomelatine are able to produce acute behavioral effects (suppression of responding) at doses lower than those for which there is any evidence of benzodiazepine-like effects. Further, the doses that suppressed responding in the present study were substantially higher than the 1–10 mg/kg doses that were active in entrainment of circadian rhythms and phase advancement in rats (Armstrong et al. 1993; Redman et al. 1995; Martinet et al. 1996).

Further evidence that melatonin does not have benzodiazepine-receptor mediated behavioral and pharmacological effects derives from antagonism studies. While substitution of triazolam for melatonin was antagonized by flumazenil, this benzodiazepine receptor antagonist did not block the discriminative stimulus effects of the training dose of melatonin (Levesque and Locke 1994). Flumazenil also failed to block the melatonin-induced potentiation of pentobarbitone sleeping time in mice (Sugden 1995) and the hypnotic and hypothermic effects of melatonin in healthy adult male humans (Nave et al. 1996).

Results of our self-administration study provide further support for the conclusions that agomelatine lacks abuse potential and has a different profile of behavioral effects than classical CNS depressant drugs. Intravenous self-administration is an animal model of the reinforcing effects of drugs in humans (Balster 1991). There is generally a strong, positive correlation between those drugs that are self-administered by laboratory animals and those that are recreationally abused by humans (Johanson and Balster 1978; Ator and Griffiths 1987). Whereas monkeys readily administered methohexital, agomelatine was not self-administered. There are no other studies we are aware of in which the reinforcing effects of a melatonin agonist have been assessed. For methohexital, injection rates above the range of rates maintained by saline were obtained at one or more doses in all four monkeys, and the inverted-U dose-effect relationship seen in most monkeys is typical of drug reinforcers. For methohexital, maximal infusion rates often exceeded 80 per 1-h session. Very different results were obtained with agomelatine. With the exception of one test dose in one monkey, substitution tests with agomelatine resulted in infusion rates that were well within or below the ranges for the tests with saline and/or the vehicle used to solubilize agomelatine. In the one test (0.1 mg/kg per infusion in M1145) where agomelatine infusion rates did exceed those obtained with vehicle, it is very unlikely that this represents a reinforcing effect because saline substitution also resulted in equally high infusion rates on one of the tests in this monkey. In addition, infusion rates steadily decreased over the last 3 days at this test dose in this monkey (data not shown), suggesting extinction of responding across consecutive days of this substitution test. Alternatively, increased infusion rates during saline tests may reflect resistance of methohexital self-administration to extinction, similar to that seen in diazepam self-administration (Grant and Johanson 1987).

It is unlikely that the lack of reinforcing effects for agomelatine demonstrated in this study is due to the failure to evaluate an adequate dosage range. Four doses of agomelatine over a 30-fold range were tested. Although infusion rates over the doses from 0.03 to 0.3 mg/kg were very similar to what were obtained with saline and vehicle, there was some evidence that the highest dose (1 mg/kg per infusion) suppressed infusion rates. In one of the monkeys, infusion rates at this dose were below the ranges obtained in this animal with vehicle and both

tests with saline. In another monkey, infusion rates at this dose were below two of the three control tests. Infusion rates below those obtained with saline or vehicle suggest that this dose may have had some behavioral effects, either direct response-rate decreasing effects or negative reinforcing effects leading to avoidance of these injections. In addition, the test dose of 1 mg/kg per infusion is quite high for self-administration studies of this type (Balster and Hayes 1993). If one were to assume that a higher test dose (e.g., 3 mg/kg per infusion) were to have had reinforcing effects had we tested it, then agomelatine would be at least 100 times less potent than methohexital, whose minimal reinforcing dose was about 0.03 mg/kg per infusion. On the other hand, no directly observable effects were noted in the monkeys after self-administration of agomelatine even though, at the highest test dose, animals often obtained in excess of 10 mg/kg in the 1-h session. No adverse health effects of agomelatine were seen in these monkeys either.

Interpretation of negative results in self-administration studies using a substitution procedure must take into account the drug used to maintain responding under baseline conditions. In general, the substitution procedure is more likely to detect reinforcing effects if the test drug is from the same pharmacological class as the baseline drug (Hoffmeister and Schlichting 1972; Bergman and Johanson 1985; Beardsley et al. 1990). In the case of agomelatine, there is little scientific basis for selecting a baseline drug that would maximize the likelihood of demonstrating reinforcing effects. As reviewed earlier, there was research suggesting that agomelatine had some acute effects similar to benzodiazepines, and use in sleep disorders is a potential clinical indication; therefore, we designed the present study under conditions that have been shown to be favorable for demonstrating reinforcing effects of benzodiazepines. Previous research, using a substitution procedure very similar to the one used here (Bergman and Johanson 1985; Johanson 1987), has shown that reinforcing effects of benzodiazepines can be obtained in rhesus monkeys maintained on barbiturate self-administration. We have confirmed this in our laboratories using methohexital-trained monkeys who self-administer diazepam and midazolam (Robert L. Balster, unpublished observations) as well as pentobarbital (Balster and Hayes 1993). Thus, the finding that agomelatine lacks reinforcing effects under conditions favorable to demonstrating the reinforcing effects of abused CNS depressant drugs is further evidence for differences between agomelatine and this class.

In summary, while melatonin-like drugs share some properties with CNS depressant drugs, they differ from this class in lacking effects in animals predictive of abuse potential. Specifically, the novel melatonin agonist agomelatine did not have reinforcing or discriminative stimulus effects similar to those of methohexital and diazepam, respectively. Melatonin itself also lacked diazepam-like discriminative stimulus effects. Taken together, these results support a prediction that agomelatine would not produce intoxication in humans similar to that produced by high doses of benzodiazepines, nor would it likely be subject to abuse.

Acknowledgements This work was performed under contract from Institut de Recherches Internationale Servier, Courbevoie, France in laboratories at Virginia Commonwealth University. The authors wish to thank David Arthur, James Hyman, and Michele Feeback for technical assistance and Elizabeth Mocaër from Servier for the supply of agomelatine and for her helpful advice on this manuscript.

References

Armstrong SM, McNulty OM, Guardiola-Lemaitre B, Redman JR (1993) Successful use of S20098 and melatonin in an animal model of delayed sleep-phase syndrome (DSPS). Pharmacol Biochem Behav 46:45–59

Ator NA, Griffiths RR (1987) Self-administration of barbiturates and benzodiazepines: a review. Pharmacol Biochem Behav 27: 391–398

Ator NA, Griffiths RR (1989) Differential generalization to pentobarbital in rats trained to discriminate lorazepam, chlordiazepoxide, diazepam, or triazolam. Psychopharmacology 98:20–30

Ator NA, Griffiths RR (1997) Selectivity in the generalization profile in baboons trained to discriminate lorazepam: benzodiazepines, barbiturates and other sedatives/anxiolytics. J Pharmacol Exp Ther 282:1442–1457

Attenburrow MEJ, Dowling BA, Sargent PA, Sharpley AL, Cowen PJ (1995) Melatonin phase advances circadian rhythm. Psychopharmacology 121:503–505

Balster RL (1990) Perception of drug effects. In: Berkley MA, Stebbins WC (eds) Comparative perception: basic mechanisms (vol 1). Wiley, New York, pp 127–154

Balster RL (1991) Drug abuse potential evaluation in animals. Br J Addict 86:1549–1558

Balster RL, Hayes BA (1993) Abuse potential evaluation of methocarbamol in rhesus monkeys. Exp Clin Psychopharmacol 1:92–100

Beardsley PM, Hayes BA, Balster RL (1990) The self-administration of MK-801 can depend upon drug-reinforcement history and its discriminative stimulus properties are phencyclidine-like in rhesus monkeys. J Pharmacol Exp Ther 252:953–959

Bergman J, Johanson CE (1985) The reinforcing properties of diazepam under several conditions in the rhesus monkey. Psychopharmacology 86:108–113

Brzezinski A (1997) Melatonin in humans. N Engl J Med 336: 186–195

Claustrat B, Le Bards D, Brun J, Thivolle P, Mallo C, Arendt J, Chazot G (1989) Plasma and brain pharmacokinetic studies in humans after intravenous administration of cold or ^{11}C labelled melatonin. In: Reiter RJ, Pang SF (eds) Advances in pineal research. Libbey, London, pp 305–310

Depreux P, Lesieur D, Mansour HA, Morgan P, Howell HE, Renard P, Caignard DH, Pfeiffer B, Delagrange P, Guardiola B, Yous S, Demarque A, Adam G, Andrieux J (1994) Synthesis and structure-activity relationships of novel naphthalenic and bioisosteric related amidic derivatives as melatonin receptor ligands. J Med Chem 37:3231–3239

de Wit H, Griffiths RR (1991) Testing the abuse liability of anxiolytic and hypnotic drugs in humans. Drug Alcohol Depend 28: 83–111

Golombek DA, Martini M, Cardinali DP (1993) Melatonin as an anxiolytic in rats: time dependence and interaction with the central GABAergic system. Eur J Pharmacol 237:231–236

Golombek DA, Pevet, P, Cardinali DP (1996) Melatonin effects on behavior: possible mediation by the central GABAergic system. Neurosci Biobehav Rev 20:403–412

Grant KA, Johanson CE (1987) Diazepam self-administration and resistance to extinction. Pharmacol Biochem Behav 28:81–86

Hagan RM, Oakley NR (1995) Melatonin comes of age? Trends Pharmacol Sci 16:81–83

Hoffmeister F, Schlichting UU (1972) Reinforcing properties of some opiates and opioids in rhesus monkeys with histories of cocaine and codeine self-administration. Psychopharmacologia 23:55–74

Holtzman SG (1990) Discriminative stimulus effects of drugs: relationship to potential for abuse. In: Adler MW, Cowan A (eds) Testing and evaluation of drugs of abuse. Wiley-Liss, New York, pp 193–210

Johanson CE (1987) Benzodiazepine self-administration in rhesus monkeys: estazolam, flurazepam and lorazepam. Pharmacol Biochem Behav 26:521–526

Johanson CE, Balster RL (1978) A summary of results of drug self-administration studies using substitution procedures in rhesus monkeys. Bull Narcot 30:43–54

Levesque TR, Locke KW (1994) Discriminative stimulus effects of melatonin in the rat. Psychopharmacology 116:167–172

Martinet L, Guardiola-Lemaitre B, Mocaer E (1996) Entrainment of circadian rhythms by S-20098, a melatonin agonist, is dose and plasma concentration dependent. Pharmacol Biochem Behav 54:713–718

Nave R, Herer P, Haimov I, Shlitner A, Lavie P (1996) Hypnotic and hypothermic effects of melatonin on daytime sleep in humans: lack of antagonism by flumazenil. Neurosci Lett 214: 123–126

Pierrefiche G, Zerbib R, Laborit H (1993) Anxiolytic activity of melatonin in mice: involvement of benzodiazepine receptors. Res Commun Chem Pathol Pharmacol 82:131–142

Redman JR, Guardiola-Lemaitre 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 118:385–390

Reppert SM, Weaver DR, Godson C (1996) Melatonin receptors step into the light: cloning and classification of subtypes. Trends Pharmacol Sci 17:100–102

Sugden D (1995) Sedative potency and 2-[^{125}I)iodomelatonin binding affinity of melatonin analogues. Psychopharmacology 117:364–370

Tallarida RJ, Murray RB (1987) Manual of pharmacologic calculations with computer programs (2nd edn). Springer, New York

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

Wiley JL, Balster RL, Martin BR (1995) Discriminative stimulus effects of anandamide in rats. Eur J Pharmacol 276:49–54

Young R, Glennon RA (1987) Stimulus properties of benzodiazepines: correlations with binding affinities, therapeutic potency, and structure activity relationships (SAR). Psychopharmacology 93:529–533