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The selective tachykinin neurokinin 1 (NK_1) receptor antagonist, GR 205,171, stereospecifically inhibits light-induced phase advances of hamster circadian activity rhythms

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Abstract

Circadian rhythms in mammals are generated by master pacemaker cells located within the suprachiasmatic nucleus of the hypothalamus. In hamsters, the suprachiasmatic nucleus contains a small collection of cells immunoreactive for substance P, the endogenous ligand of tachykinin neurokinin 1 (NK_1) receptors. In addition, two other nuclei which form part of the circadian system, the intergeniculate leaflet of the thalamus and the raphe nuclei, also contain fibers and/or cell bodies immunoreactive for substance P. In light of these observations, we evaluated the influence of the selective tachykinin NK_1 receptor antagonist, GR 205,171, upon circadian activity rhythms in the hamster. Systemic injection of GR 205,171 dose-dependently (2.5–40.0 mg/kg, i.p.) inhibited light-induced phase advances in hamster circadian wheel running activity rhythms by approximately 50%. In contrast, GR 226,206, the less active enantiomer of GR 205,171, failed to affect light-induced phase advances. In addition, we examined the potential ability of GR 205,171 to induce non-photic phase shifts in hamster wheel running rhythms when injected at mid-day to late night circadian times. However, GR 205,171 (40 mg/kg) did not elicit non-photic phase shifts at these times indicating that tachykinin NK_1 receptor antagonists are only effective when a light stimulus is applied to the pacemaker. Although GR 205,171 may, in theory, activate several sites within the circadian system, we suggest that GR 205,171 acts in the raphe nuclei to increase inhibitory serotonergic input to pacemaker cells in the suprachiasmatic nuclei, thereby suppressing photic modulation of the pacemaker. These findings have important implications for the use of tachykinin NK_1 receptor antagonists in the treatment of depression and other central nervous system disorders.

Keywords: Circadian; Suprachiasmatic; Neurokinin; Raphe; Depression

1. Introduction

The primary pacemaker for circadian rhythms in mammals is located in the suprachiasmatic nuclei at the base of the hypothalamus (Ralph et al., 1990). The suprachiasmatic nuclei are innervated by retinal axons arriving via the retinohypothalamic tract, and information regarding the light/dark cycle conveyed by this pathway is used by the pacemaker to adjust the timing of the clock to changes in day-length during the year. Glutamate is the primary transmitter of the retinal afferents to the suprachiasmatic nuclei (Ebling, 1996), though pituitary adenylate cyclase activating peptide is well-established to be co-localized with glutamate in retinohypothalamic tract terminals (Chen et al., 1999; Gillette and Mitchell, 2002).

As regards other neuropeptides, there is considerable interest in the potential significance of substance P, the endogenous ligand of tachykinin NK_1 receptors. In fact, there is some disagreement as to whether substance P is also contained within the retinohypothalamic tract terminals, and this appears to differ in a species-dependent fashion. Thus, several reports suggest that substance P is found in the retinohypothalamic tract of rats (Mikkelsen and Larsen, 1993; Piggins et al., 2001), whereas others suggest that it is *not* (Hannibal and Fahrenkrug, 2002; Hartwich et al., 1994; Otori et al., 1993). There is also a report suggesting that substance P is contained within the human retinohypothalamic tract (Moore and Speh, 1994). However, there is a consensus that substance P is *not* part of the

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retinohypothalamic tract in hamsters or mice (Hartwich et al., 1994; Piggins et al., 2001).

The suprachiasmatic nuclei also receive information from the intergeniculate leaflet of the thalamus via the geniculohypothalamic tract which contains γ-aminobutyric acid (GABA) and neuropeptide Y as the primary transmitters, while enkephalin is also found in the hamster (Harrington et al., 1985; Morin et al., 1992). The intergeniculate leaflets of rat, hamster and mice all contain axons immunoreactive to substance P and tachykinin NK_1 receptors are also localized in this structure (Morin et al., 1992; Piggins et al., 2001). In addition to the intergeniculate leaflet, the suprachiasmatic nuclei are innervated by brainstem raphe nuclei, in particular the median raphe in hamsters (Meyer-Bernstein and Morin, 1996). Although raphe input to the suprachiasmatic nuclei is primarily serotonergic, substance P and tachykinin NK_1 receptors are found within the raphe nuclei (Leger et al., 2002; Meyer-Bernstein and Morin, 1996), and are believed to modulate the output of these nuclei (see Discussion). Finally, there is a small group of substance P immunoreactive cells located within the central region of the suprachiasmatic nuclei in the hamster, rat and primate, but not the mouse (Hartwich et al., 1994; Mick et al., 1992; Mikkelsen and Larsen, 1993; Morin et al., 1992; Piggins et al., 2001; Reuss et al., 1994). Therefore, in the hamster, substance P could influence circadian rhythmicity by acting in the suprachiasmatic nuclei, intergeniculate leaflet, or raphe nuclei.

The potential influence of substance P upon circadian rhythms has been investigated in both rats and hamsters. In the rat hypothalamic slice in vitro, substance P shifts the rhythm of spontaneous neuronal firing in a manner analogous to that of light (Shibata et al., 1992). Also in the slice, optic nerve-evoked phase shifts in suprachiasmatic nuclei neuronal firing are inhibited by tachykinin NK_1 receptor antagonists while exogenously applied substance P elicits shifts in neuronal firing frequencies (Hamada et al., 1999; Kim et al., 2001). The phase-shifting effects of substance P are thought to result from an increase in glutamate release from the rat retinohypothalamic tract (Hamada et al., 1999; Kim et al., 1999, 2001). In the hamster, tachykinin NK_1 receptor antagonists block light-induced c-*fos* expression in the suprachiasmatic nuclei (Abe et al., 1996), as well as phase advances in circadian activity rhythms (Challet et al., 1998, 2001), following i.c.v. or systemic injections, respectively. Thus, the site of action for tachykinin NK_1 receptor antagonists in the hamster has not been definitively linked to the suprachiasmatic nuclei, but may be acting elsewhere in the circadian system. However, in both the rat and hamster, substance P mimics the effect of light on the circadian pacemaker.

Pharmacological studies of circadian rhythms are important inasmuch as novel drugs are needed for the normalization of dysfunctional circadian rhythms in central nervous system disorders such as Alzheimer's and Parkinson's disease, insomnia and depression (Harper et al., 2001; Reid et al., 2004). Moreover, irregularities in human circadian rhythms have been specifically implicated, but not unequivocally confirmed, in the aetiology of Seasonal Affective Disorder (Burgess et al., 2004; Koorengevel et al., 2003; Magnusson and Boivin, 2003; Rosenthal et al., 1984). As regards potential therapeutic mechanisms, an evaluation of the influence of tachykinin NK_1 receptor antagonists upon circadian rhythms is of special interest in view of their current development for the treatment of major depression and anxiety disorders (Duffy, 2004; Millan, 2003; Rupniak and Kramer, 1999; Rupniak et al., 2001).

In the present study we examined the ability of the highly selective tachykinin NK_1 receptor antagonist GR 205,171 to modulate both light-induced and non-photic phase shifts in hamster circadian activity rhythms. GR 205,171 is the most selective and best characterized tachykinin NK_1 receptor antagonist designed to date (Gardner et al., 1996; Millan et al., 2001; Saria, 1999). To confirm the specificity of its potential actions, we examined in parallel the actions of its less active stereoiosmer, GR 206,226. In addition, potential non-photic activity of tachykinin NK_1 receptor antagonists acting at times other than late in the mid-day is reported here for the first time. Finally, recent evidence from other laboratories allows us to propose a truly novel site and mechanism of action for the phase-modulating activity of tachykinin NK_1 receptor antagonists, the raphe nuclei.

2. Materials and methods

Young (80 g) male Syrian hamsters (*Mesocricetus auratus*) were purchased from Charles River Laboratories (Kingston, NY) and maintained in a 14 : 10 h light : dark cycle for at least two weeks prior to use in experiments. Food and water were provided ad libitum. Hamsters were transferred to individual cages equipped with small running wheels (19 cm diameter) and placed in conditions of constant darkness for the duration of each experiment (about three weeks in constant darkness). Wheel revolutions were identified using magnets and magnetic switches attached to the running wheel and cage lid, respectively, and recorded using hardware and software from Actimetrics (Wilmette, IL, USA) and Matlab (The MathWorks, Natick, MA, USA). All experiments were approved by the Institutional Animal Care and Use Committee and conform to the European Community guidelines for the use of experimental animals.

The time of wheel running onset by hamsters in constant darkness each day is identified as circadian time 12 (12 h of a 24 h day). For each experiment day, circadian time 12 was determined by fitting a line through circadian time 12 of the previous five days, and extrapolating for circadian time 12 the next day. Phase shifts in activity onsets were determined by fitting a line through days five–ten post-experiment, and then comparing the intercept difference between the slopes of the pre- and post-experiment lines on the day of the experiment.

For photic experiments, after the hamsters had been in constant darkness for approximately ten days, the hamsters were removed from their cages in constant darkness under dim red light (<1 lx), weighed, and injected with either drug or vehicle and returned to their home cage in constant darkness. Forty-five minutes later at circadian time 19, hamsters were

removed again from constant darkness and exposed to a 10-min pulse of 20 lx white light (described in Tierno et al., 2002) and then returned to constant darkness for the duration of the experiment (approximately ten more days).

For non-photic experiments, hamsters were removed from their home cage at various circadian times after about ten days in constant darkness weighed and injected with either drug or vehicle under dim red light, and returned to their home cage for the duration of the experiment (about ten days more). The hamsters were out of their home cage for less than 1 min.

GR 205,171 is 2-methoxy-5-(5-trifluoromethyl-tetrazol-1-yl)-benzyl]-(2*S*-phenyl-piperidin-3*S*-yl)-amine 2HCl and GR 226,206 is 2-methoxy-5-(5-trifluoromethyl-tetrazol-1-yl)-benzyl]-(2*S*-phenyl-piperidin-3*R*-yl)-amine 2HCl. Both drugs were synthesized by Servier chemists. They were dissolved in sterile water and injected by the intraperitoneal (i.p.) route. Doses are expressed as milligram per kilogram base. Analysis of variance (ANOVA) and Tukey statistical tests were used for the photic experiments. Paired *t*-test was used for statistical analysis of the non-photic experiment. All data are expressed as the means ± standard errors of the means (S.E.M.).

3. Results

3.1. Photic experiments: light-induced phase shifts of hamster activity rhythms

Hamsters exposed to a 10-min, 20 lx white light pulse at circadian time 19 following vehicle injections phase advanced their circadian wheel running activity rhythms by 1.08 ± 0.1 h (mean ± S.E.M., Fig. 1). The selective tachykinin NK_1 receptor antagonist, GR 205,171, dose-dependently inhibited the phase advance evoked by the light pulse by a maximum of ca. 50% at the highest dose tested 0.57 ± 0.07 h, 40 mg/kg GR 205,171 (Fig. 1). One-way ANOVA revealed this effect to be highly significant (one-way ANOVA, $F_{3,19} = 7.48$, $P = 0.0016$). A post hoc Tukey confirmed that the inhibitory effects of 20 and 40 mg/kg GR 205,171 upon the phase advance were significantly different from vehicle ($P < 0.01$). There were no significant differences between the lengths of circadian activity periods between vehicle animals and those receiving 40 mg/kg GR 205,171 prior to the day of injections. Period for vehicle = 24.02 ± 0.04 h and period for 40 mg/kg GR 205,171 = 24.02 ± 0.01 h. In addition, the total

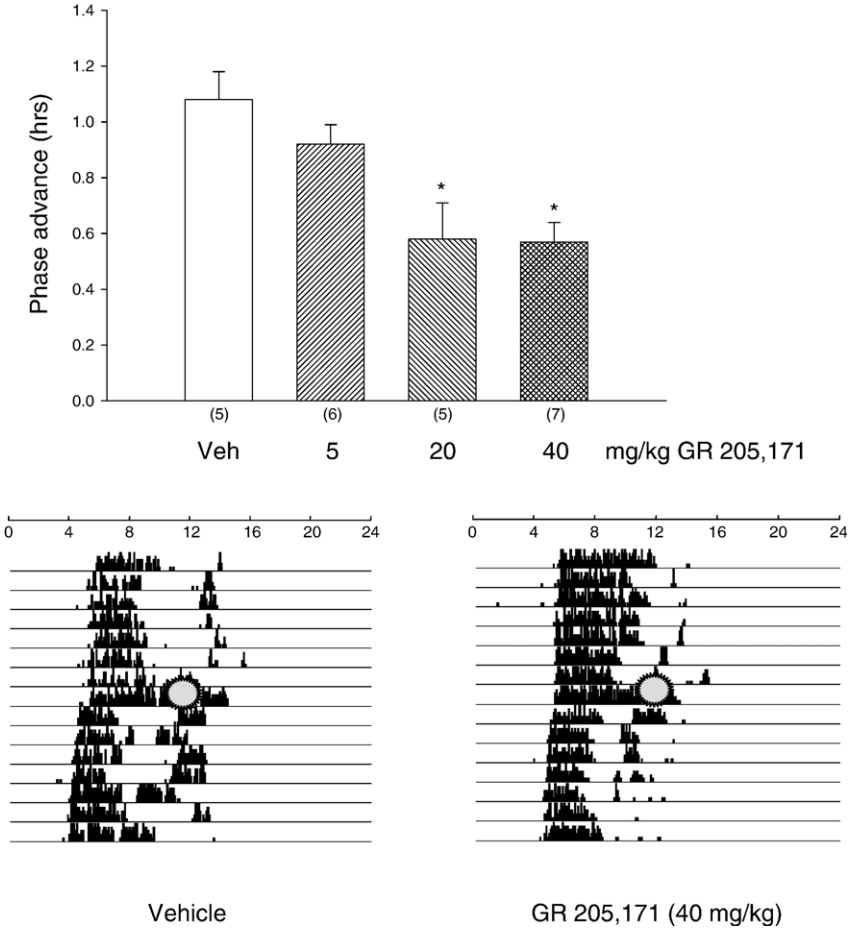

Fig. 1. Inhibition by the selective tachykinin NK_1 receptor antagonist, GR 205,171, of light-induced phase shifts in hamster circadian wheel running activity rhythms. Top: Dose response relationship for or the actions of GR 205,171. The number of hamsters used for each dose is indicated in parentheses below the bars. Veh = vehicle. *$P < 0.01$ from vehicle. Bottom: Representative actograms for data shown in the above panel. The top scale is a 24 h day. Wheel running activity for a total of 15 days is shown, each row corresponds to one day. Vertical bars in each row indicate the relative number of wheel revolutions collected at 10-min intervals. The stars indicate the time of light exposure.

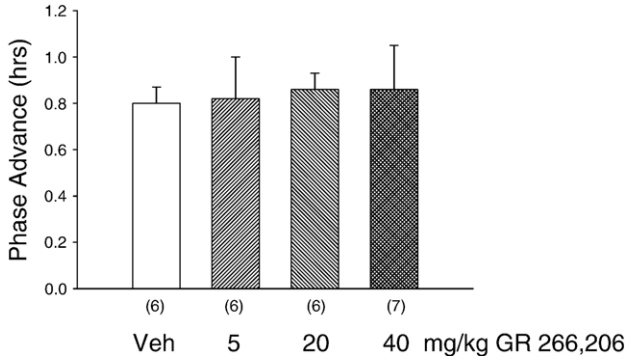

Fig. 2. Lack of inhibition by GR 226,206, the less active enantiomer of GR 205,171, of light-induced phase shifts in hamster circadian wheel running activity rhythms. GR 226,206 did not affect light-induced phase shifts of hamster circadian activity rhythms when administered at the same doses as GR 205,171. The number of hamsters tested at each dose is indicated in parentheses below the bars.

number of wheel revolutions on the day of the light exposure was also not significantly different between the two groups: vehicle = 7950 ± 1428 revolutions versus 9801 ± 1641 revolutions for 40 mg/kg GR 205,171 (one-way ANOVA, $F_{1,10} = 0.65$, $P = 0.44$).

In the experiment in which actions of the less active enantiomer, GR 226,206, were tested, the light-induced phase shift following vehicle administration was 0.8 ± 0.07 h (Fig. 2). GR 226,206 was administered over the same dose-range as employed for GR 205,171, but failed to exert a significant effect upon light-induced phase shifts at any doses tested (one-way ANOVA $F_{3,21} = 0.03$, $P = 0.98$, Fig. 2). The action of GR 205,171 is, thus, exerted specifically.

3.2. Non-photic experiments: drug-induced phase shifts of hamster activity rhythms

The maximally effective dose of GR 205,171 in the photic experiments was also tested for its ability to phase shift hamster circadian activity rhythms in the *absence* of light (referred to as non-photic shifts). GR 205,171 (40 mg/kg) was tested at circadian times 6, 10, 14, 18, and 22, times which cover the mid-day to late night period of the circadian cycle. GR 205,171 failed to shift the circadian wheel running rhythms by more than 10 min at any circadian time tested, and paired *t*-tests conducted at each time indicated that there were no significant differences between vehicle and GR 205,171 injections at any circadian time (Fig. 3).

4. Discussion

4.1. Stereospecific inhibition of light-induced phase shifts by GR 205,171

In this report, we demonstrate that the tachykinin NK_1 receptor antagonist, GR 205,171 dose-dependently inhibits light-induced phase advances of hamster circadian activity rhythms. In contrast to effects seen with light, it had no influence on the circadian phase of wheel running activity when injected without concomitant light pulses (non-photic). Therefore, these results show that selective blockade of tachykinin NK_1 receptors modulates photic input to the suprachiasmatic nuclei circadian pacemaker. Several arguments support a specific role of tachykinin NK_1 receptors in the actions of GR 205,171.

First, the action of GR 205,171 was exerted stereospecifically inasmuch as GR 226,206, its less active isomer (Cumberbatch et al., 1998; Gardner et al., 1996; Millan et al., 2001, 2002), did *not* significantly modify the influence of light pulses upon circadian rhythms. Furthermore, GR 205,171 is an exceptionally selective ligand for tachykinin NK_1 receptors with no known activities at other classes of binding site (Gardner et al., 1996; Millan et al., 2001 and unpub. obs; Saria, 1999). *Second*, the actions of GR 205,171 were exerted dose-dependently and were specific to conditions of light pulses. *Third*, though stereoselectivity of effects was not evaluated, and only a single dose was employed, two further tachykinin NK_1 receptor antagonists, L 760,735 and R 116,301, suppressed the phase advances in circadian rhythms provoked by light in hamsters (Challet et al., 1998, 2001). *Fourth*, doses of GR 205,171 active in the present work coincide well with those effective in other functional models of the blockade of central tachykinin NK_1 receptors in rats: notably, inhibition of marble-burying behaviour and induction of frontocortical release of noradrenaline in rodents, actions related to the antidepressant properties of tachykinin NK_1 receptor antagonists (Blier et al., 2004; Clayton et al., 1997; Cumberbatch et al., 1998; Maubach et al., 2002; McAllister and Prat, 1998; Millan et al., 2001, 2002). *Finally*, the active dose-range of GR 205,171 herein corresponds well to ex vivo binding studies suggesting that doses of ca. 10.0 mg/kg of

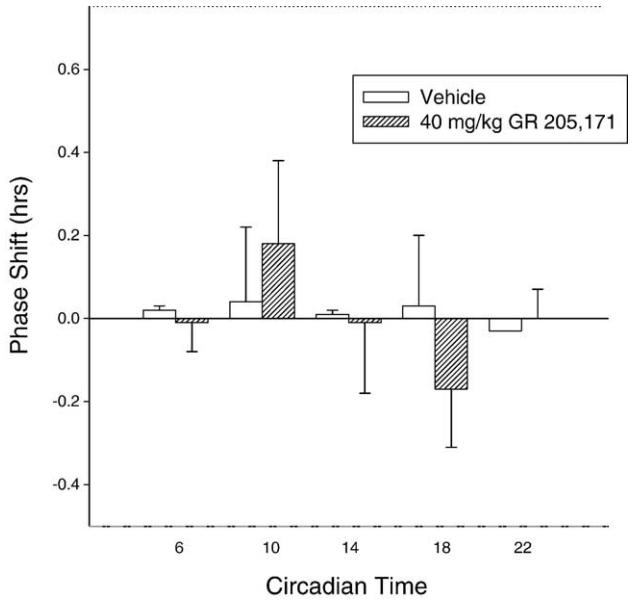

Fig. 3. Lack of influence of the selective tachykinin NK_1 receptor antagonist, GR 205,171, upon non-photic phase shifts in circadian activity rhythms. Non-photic phase shifts were measured following injection of vehicle or GR 205,171 at five times representing mid-day to late night times. $n = 5$ for each condition at circadian times 14, 18, 22; $n = 4$ for circadian time 6, and $n = 3$ for circadian time 10.

GR 205,171 are required to substantially occupy central tachykinin NK_1 receptor sites (Hutson et al., 2004).

In line with the foregoing comment, it was mentioned in a previous report that a selective tachykinin NK_1 receptor antagonist which does not pass the blood–brain barrier fails to modify the phase-advancing effect of a light pulse (Challet et al., 1998). This observation further suggests that GR 205,171 is acting centrally to exert its inhibitory actions. However, the circadian system in hamsters is composed of the retina, the suprachiasmatic nuclei, the intergeniculate leaflet and the dorsal and median raphe nuclei. Since we used systemic i.p. injections in this study, GR 205,171 could be acting at any of these locations, and each possibility is discussed in turn below.

4.2. Potential actions at tachykinin NK_1 receptors in the retina

There is a single report that indicates substance P is located in the hamster retina, (Li et al., 1999; Mikkelsen and Larsen, 1993; Piggins et al., 2001) and a small number of retinal ganglion cells immunoreactive to substance P have also been identified in the hamster (Li et al., 1999). However, substance P has not been found in retinohypothalamic tract fibers synapsing within the suprachiasmatic nuclei, so the substance P immunoreactive retinal ganglion cells found in hamster retina may be projecting elsewhere in the brain. In the above-mentioned report with the tachykinin NK_1 receptor antagonists, L 760,735 and R 116,301, the authors also administered them after the phase-shifting light pulse, and, if effective, this would eliminate the retina as their site of action. Under these conditions, L 760,735 and R 116,301 failed to inhibit light-induced phase shifts, so in that experiment the retina cannot be ruled out as a site of action for tachykinin NK_1 receptor antagonists (Challet et al., 1998, 2001). However, in the same report (Challet et al., 1998), a tachykinin NK_1 receptor antagonist that cannot pass the blood–brain barrier failed to modulate light-induced phase shifts, and this argues against a peripheral (retinal) site of action for tachykinin NK_1 receptor antagonist activity. It is probably safe, however, to rule out pupil constriction as a mechanism of activity for GR 205,171. Thus, substance P induces miosis in the eye, an effect blocked by tachykinin NK_1 receptor antagonists (Alessandri et al., 1991; Andersson and Almegard, 1993; Hay et al., 2002), suggesting that they would be expected, if anything, to enhance the amount of light entering the retina. Therefore, it is unlikely that the effects of GR 205,171 on phase advances reflect pupillary constriction.

4.3. Potential actions at tachykinin NK_1 receptors in the suprachiasmatic nuclei

Both the rat and, more clearly, the hamster contain a small collection of substance P immunoreactive cells within the suprachiasmatic nuclei (Hartwich et al., 1994; Mikkelsen and Larsen, 1993; Morin et al., 1992; Piggins et al., 2001; Reuss et al., 1994). Substance P applied in vitro directly to the rat suprachiasmatic nuclei shifts the rhythm of spontaneously firing neurons in a manner similar to the effects of light in vivo (Shibata et al., 1992), and optic nerve-evoked shifts in neuronal firing rhythms in the rat suprachiasmatic nuclei are inhibited by tachykinin NK_1 receptor antagonists in vitro (Kim et al., 1999). Further, substance P increases the release of glutamate within the rat suprachiasmatic nuclei in vitro (Hamada et al., 1999; Shirakawa and Moore, 1994). In the hamster hypothalamic slice in vitro, ionophoretically applied substance P both excites and inhibits the spontaneously firing neurons in the lateral and medial aspects of the ventral suprachiasmatic nuclei (Piggins et al., 1995). Taken together, these reports indicate that the suprachiasmatic nuclei could be the site of action for GR 205,171 in the hamster. However, when substance P was injected directly into the hamster suprachiasmatic nuclei in vivo, it had little effect (Piggins and Rusak, 1997). In addition, tachykinin NK_1 receptors are not found in the hamster suprachiasmatic nuclei (Piggins et al., 2001). Therefore, these studies by Piggins et al. (2001) and Piggins and Rusak (1997) diminish the likelihood that the suprachiasmatic nuclei are a site of GR 205,171 activity.

4.4. Potential actions at tachykinin NK_1 receptors in the intergeniculate leaflet

The intergeniculate leaflet of the thalamus contains substance P immunoreactive axons and tachykinin NK_1 receptors both in hamsters and in rats (Morin et al., 1992; Piggins et al., 2001). Neuropeptide Y and enkephalins in the hamster geniculohypothalamic tract transfer information from the intergeniculate leaflet to the suprachiasmatic nuclei. Light-induced phase shifts in hamster circadian activity rhythms are inhibited by agonists to both neuropeptide Y and enkephalin (δ-opioid) receptors (Tierno et al., 2002; Weber and Rea, 1997). In addition, agonists for neuropeptide Y and δ-opioid receptors induce non-photic phase shifts in hamster circadian activity rhythms when injected in the mid-late day (Byku and Gannon, 2000; Yannielli and Harrington, 2004). If GR 205,171 was active in the intergeniculate leaflet, then it should mimic the effects of neuropeptide Y and enkephalin in eliciting non-photic phase shifts in hamster activity rhythms during the day, which it does not (Fig. 3). Therefore, even though the mediation of photic and non-photic effects from the intergeniculate leaflet to the suprachiasmatic nuclei could occur through separate pathways, it is more likely that the intergeniculate leaflet is not the site of action for GR 205,171 in the hamster.

4.5. Potential actions at tachykinin NK_1 receptors in the median raphe

The median raphe nucleus is the primary source for serotonergic afferents synapsing within the hamster suprachiasmatic nuclei (Meyer-Bernstein and Morin, 1996). There are also considerable interactions between the dorsal and median raphe nuclei, and the former project to the intergeniculate leaflet in the hamster (Tischler and Morin, 2003). Therefore, activation of either the median or dorsal raphe nuclei can modulate input to the hamster suprachiasmatic nuclei. Exogenously applied serotonergic agonists inhibit light-induced phase shifts in hamster circadian activity rhythms, and the raphe projections to the suprachiasmatic nuclei are believed

to inhibit the effects of light on the circadian pacemaker (Morin, 1999). Recently, it was reported that tachykinin NK_1 receptor antagonists increase the firing rate of raphe serotonergic neurons and the activity of these neurons is also enhanced in mice genetically deprived of tachykinin NK_1 receptors (Blier et al., 2004; Guiard et al., 2004; Millan, 2003; Santarelli et al., 2002; Valentino et al., 2003). In a complex arrangement within raphe nuclei, substance P may activate glutamatergic terminals which excite serotonergic perikarya: locally released serotonin then activates serotonin$_{1A}$ autoreceptors leading to an inhibition of the firing of serotonergic neurons (Liu et al., 2002; Valentino et al., 2003). Tachykinin NK_1 antagonists such as GR 205,171 would prevent this auto-inhibition of raphe neurons, thus enhancing inhibitory raphe input to the suprachiasmatic nuclei upon exposure to a light pulse. This model would explain the inhibitory activity of GR 205,171 in this study. In addition, there is evidence that modulation of the circadian phase by raphe is *not* exerted tonically and that this control is not active in the absence of light (Morin, 1999). Accordingly, GR 205,171 does not elicit *non*-photic phase shifts. This model may also explain the finding that L 760,735 and R 116,301 were only able to induce phase shifts for hamsters housed in constant light, but not constant dark (Challet et al., 1998, 2001).

4.6. Summary and conclusions

Altogether, we propose that tachykinin NK_1 receptor antagonists inhibit light-induced phase advances in hamster circadian activity rhythms by enhancing inhibitory serotonergic input from the raphe to the suprachiasmatic nuclei pacemaker. However, this remains to be demonstrated by measures of serotonin release in the suprachiasmatic nuclei of hamsters. Further, several studies suggested that the facilitatory influence of tachykinin NK_1 receptor blockade upon serotonergic neurons is most marked in the *long*- rather than short-term. Inasmuch as the present work was undertaken with acute administration of GR 205,171, its effects upon chronic treatment would be of interest to evaluate. Tachykinin NK_1 receptor antagonists are under development as novel anxiolytic and antidepressive agents (Blier et al., 2004, Duffy, 2004; Hargreaves, 2002; Kramer et al., 1998, 2004; Millan, 2003; Rupniak and Kramer, 1999). Abnormalities in circadian rhythmicity are known to be associated with, and perhaps contribute to the pathogenesis of, affective disorders (Introduction). It is, thus, critical to evaluate the effects of putative antidepressant agents upon circadian rhythms. The present report suggests that tachykinin NK_1 receptor antagonists may be valuable in controlling photic input to the circadian pacemaker in humans and, possibly, helping to resynchronise perturbed rhythms in depressed subjects. More generally, tachykinin NK_1 receptor antagonists may be useful where individuals are subjected to rapidly changing schedules of light, such as those which occur during shift work or transmeridian jet travel: under such circumstances, it may be beneficial to maintain the original phase of the body's circadian rhythms.

Acknowledgements

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