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Evening melatonin and bright light administration induce additive phase shifts in dim light melatonin onset

Abstract: In healthy young men, administration of a single light pulse (5000 lux for 3 hr) or a single melatonin pill (5 mg) at 20:40 hr under controlled constant routine conditions of < 10 lux, yielded a phase delay and a phase advance, respectively, in the circadian marker of dim light melatonin onset 24 hr later. Phase shifts after combining the two interventions were additive. Melatonin suppression is not necessary for a phase shift by light, and melatonin is not a 'weak' Zeitgeber relative to bright light when ambient lighting is strictly controlled.

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Introduction

Evening light exposure (even at relatively low levels) suppresses the onset of melatonin secretion and leads to a phase delay of the circadian pacemaker in humans [1–3]. Yet it is not clear whether melatonin suppression is necessary to induce this phase delay, as phase shifts with light occur during the day when melatonin is not secreted [4, 5]. We thus administered evening light in the presence or absence of exogenous melatonin. Evening light protracted, melatonin accelerated the evening decline in core body temperature (CBT), but concurrent melatonin administration did not block the light-induced phase delay in CBT measured on the following day [6]. For unequivocal interpretation of these findings it was necessary to measure the most stringent marker of the circadian pacemaker in humans, the timing of endogenous melatonin secretion onset.

Methods

In a placebo-controlled 4-week within-subject crossover design, nine healthy young men were studied for 2 days each time in the laboratory. During the morning they were seated under room light, from 14–24 hr they lay supine in bed in a modified constant routine protocol (< 10 lux, 22°C) followed by nocturnal sleep (for methodological details see [6, 7]). Treatments were: 5 mg melatonin or placebo at 20:40 hr, with or without a subsequent 3 hr light pulse (5000 lux) from 21–24 hr. All groups received placebo at the same time on the second day. Saliva was collected at 30-min intervals and melatonin assayed by RIA (Bühlmann Laboratories, Allschwil, Switzerland) with analytical least detectable dose 0.15 pg/mL and functional least detectable dose 0.65 pg/mL [8]. The time of dim light melatonin onset (DLMO), a validated circadian marker [9], was defined by a 3-pg/mL threshold crossing. One subject's results could not be used for technical reasons, the second had missing samples. Data for the remaining seven subjects were analysed with two-way ANOVA for repeated measures.

Results

On the treatment day (Fig. 1, left panel), melatonin secretion was low during the early evening, and all groups showed a similar timing of the melatonin rise. After exogenous melatonin administration at 20:40 hr, maximum peak values were observed 20 min later, followed by an exponential decline. The 3-hr light pulse subsequent to melatonin intake did not significantly affect these exogenous melatonin kinetics. However, the light pulse subsequent to placebo intake immediately suppressed endogenous melatonin levels, which remained low for the remainder of the measurement period.

On the post-treatment day (Fig. 1, right panel) melatonin profiles 24 hr later differed. The two-way ANOVA with DLMO times yielded significant main effects for light [$F_{(1,6)} = 16.02$, $P < 0.007$] and melatonin [$F_{(1,6)} = 7.50$, $P < 0.034$] but no significant light × melatonin interaction term [$F_{(1,6)} = 0.90$, n.s.]. These main effects yielded a delay in DLMO of 41 ± 10 (SEM) min with light and an advance of 24 ± 8 min with melatonin. The combination of evening melatonin and light were additive in their phase shifting effects, leading to a curve not significantly different

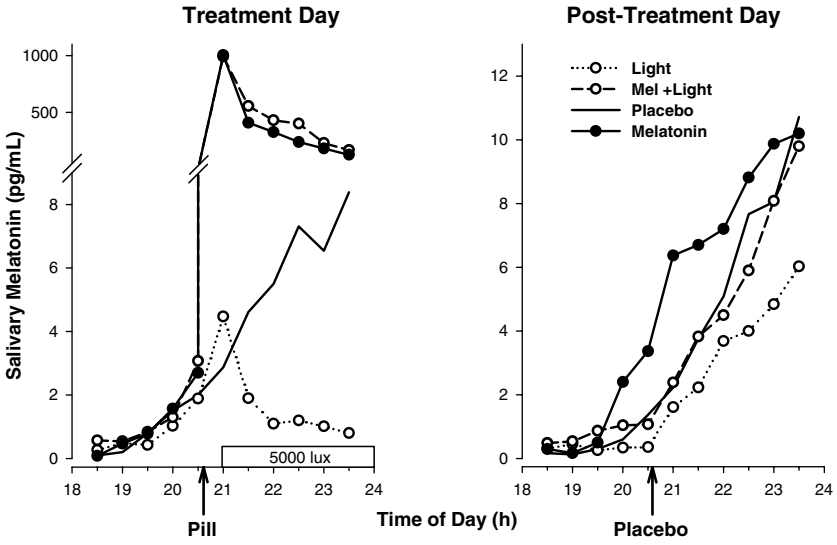

Fig. 1. Melatonin profiles were averaged over each time point for the four treatment groups on the treatment (left) and post-treatment (right) day (n = 7; for clarity, SEMs are not shown; pill at 20:40 hr on both days). Melatonin administration (5 mg) yielded pharmacological levels, light could suppress endogenous secretion. To measure phase shifts 24 hr later, two-way ANOVA for repeated measures was carried out with the individually determined dim light melatonin onset values (see text for statistics).

from placebo. Note, that for statistics we used individual DLMO determinations on the time axis; the figure presents, in contrast, mean values (pg/mL) of the raw data for each time point.

Discussion

We previously showed that the rhythms of CBT, subjective sleepiness, and sleep onset, were phase delayed by evening light, independent of whether melatonin was co-administered or not [6, 10]. The DLMO results confirm this finding and show that suppression of melatonin does not appear to be a prerequisite for phase shifting the human circadian pacemaker.

Although the correlation between the two main phase markers of melatonin and CBT rhythms was very high ($r = 0.88$, n = 32, $P < 0.0001$), only DLMO was sensitive enough to show that melatonin significantly advanced its own secretion. These new findings reveal that a single melatonin administration can phase advance the human circadian pacemaker later than previously thought (3 hr before bedtime, at the time of endogenous melatonin onset). According to the most-cited phase–response curve (PRC) to melatonin, no phase shift should have resulted from melatonin administration at CT14 [11]. However, this PRC was not carried out under such low lighting or constant routine conditions as ours, and used repeated administrations for 4 days. The PRC to a single 3-hr melatonin infusion was able to measure phase advances at this time of day [12].

Given the extremely controlled conditions in our experiment, it is possible to compare the phase-shifting ability of these two Zeitgebers, administered as a 'single pulse' in a specific dose (3 hr 5000 lux light versus 5 mg melatonin). Melatonin has been considered a 'weak' Zeitgeber (by *ca.* 10 orders of magnitude [11]), yet the advance induced by this single administration was already half that of the light pulse. At a more optimum time on the PRC (18 hr) a single melatonin administration (same dose, same constant routine conditions) advanced phase by 44 min (Table 3 in [7]). This is of the same order of magnitude as found here for the evening light pulse. It is clear, that under normal ambient conditions light has by far the most prominent effect (summation of photons over the waking day, 'masking' of melatonin at night), but at the neurobiological level, the pineal hormone may be considered to have near Zeitgeber equivalence.

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