Temporal dynamics of late-night photic stimulation of the human circadian timing system

Jamie M. Zeitzer, Sat Bir S. Khalsa, Diane B. Boivin, Jeanne F. Duffy, Theresa L. Shanahan, Richard E. Kronauer, and Charles A. Czeisler. Temporal dynamics of late-night photic stimulation of the human circadian timing system. Am J Physiol Regul Integr Comp Physiol 289: R839 –R844, 2005. First published May 12, 2005; doi:10.1152/ajpregu.00232.2005.—The light-dark cycle is the primary synchronizing factor that keeps the internal circadian pacemaker appropriately aligned with the environmental 24-h day. Although it is known that ocular light exposure can effectively shift the human circadian pacemaker and do so in an intensity-dependent manner, the curve that describes the relationship between light intensity and pacemaker response has not been fully characterized for light exposure in the late biological night. We exposed subjects to 3 consecutive days of 5 h of experimental light, centered 1.5 h after the timing of the fitted minimum of core body temperature, and show that such light can phase advance shift the human circadian pacemaker in an intensity-dependent manner, with a logistic model best describing the relationship between light intensity and phase shift. A similar sigmoidal relationship is also observed between light intensity and the suppression of plasma melatonin concentrations that occurs during the experimental light exposure. As with a simpler, 1-day light exposure during the early biological night, our data indicate that the human circadian pacemaker is highly sensitive even to typical room light intensities during the late biological night, with ~100 lux evoking half of the effects observed with light 10 times as bright.

Light has been demonstrated to be the preeminent Zeitgeber (Gm., time cue) across the phylogenetic chart (13, 27). In humans, bright light administered during the early biological night evokes delays in the timing of the circadian system, whereas bright light administered during the late biological night evokes advances in its timing (20, 21). Although early experiments on the nature of the photic resetting effect in humans used exposure to very bright light (8, 11, 14), later experiments indicated that the human circadian pacemaker is sensitive to lower intensities of light (2, 3, 12, 33, 36). Recently, it has been demonstrated that the dose-response relationship of the human circadian pacemaker to light administered outside the critical zone (19, 34), during the early subjective night, can be described best by a logistic function, as observed in the output measures of melatonin phase shift, melatonin suppression, and acute enhancement of alertness (7, 36). It has been reported, however, that the dose-response relationship to light administered during the late subjective night is nonlinear and seemed to be consistent with a cube-root compression of illuminance (2, 3), as has been observed for human judgment of luminance values in psychophysical experiments (31). Although there were limited data <180 lux (typical indoor room illumination), it was recognized that this postulated cube-root relationship could not account for responses observed to light <180 lux (18). As a cube-root function can predict very different responses to light than a logistic function, it was our intent to examine the effects of light administered during the late biological night and determine the model that best fit the response of the human circadian pacemaker, as observed through shifts in the timing of the rhythm of plasma melatonin and acute light-induced suppression of plasma melatonin concentrations.

MATERIALS AND METHODS

We studied 48 healthy, male subjects, aged 18–30 yr [23 ± 3.4 yr (mean ± SD)]. The subjects’ health was confirmed by history and physical, chemistries, interview, and questionnaires. For at least 2 wk before entering the lab, all subjects maintained a regular sleep-wake (8 h sleep/16 h wake) schedule, as confirmed by wrist actigraphy (Ambulatory Monitoring, Ardsley, NY) and sleep logs. During this time, subjects also refrained from alcohol, caffeine, and nicotine, as well as all other prescription, nonprescription, and recreational drugs; this was confirmed by urinary toxicological screening on the day of entry into the lab. All subjects gave informed written consent; all experimental procedures were carried out in accordance with the principles of the Declaration of Helsinki and approved by the Brigham and Women’s Hospital human research committee.

Protocol. Upon entry to the lab, each subject spent at least 11 days residing singly in a laboratory suite that was free from time cues (i.e., no radio, television, clock, or other time-keeping device or any contact with nonlaboratory members). No napping or strenuous exercise was allowed during the study period. Throughout the study, subjects had their core body temperature continuously recorded via a rectal thermistor (YSI, Yellow Springs, OH) and stored as the average of 1-min intervals. In addition, beginning on day 2, blood samples were taken 1–3 times per hour via an indwelling, intravenous forearm catheter. Collected samples were immediately spun in a refrigerated centrifuge (2–4°C), the plasma was decanted, frozen, and later assayed for melatonin concentration by radioimmunoassay (assay sensitivity of 2.5 pg/ml; intra-assay and interassay coefficients of variation, 8 and 13%, respectively; DiagnosTech, Osceola, WI). The first 3 days were spent as a baseline, with the subject rising at his typical wake time and going to bed 16 h later. The midpoint of these baseline sleep episodes was calculated by averaging the midpoints of the previous week of at-home sleep episodes. Ambient illumination during the 16 h of wakefulness did not exceed 150 lux, and during the 8 h of sleep, illumination was kept to <0.03 lux. All light measurements were

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
taken in the horizontal angle of gaze by an IL1400 or IL1700 research photometer fitted with a photopic and a cosine filter (International Light, Newburyport, MA).

Upon awakening on day 4, subjects began a constant routine, a procedure in which the subject remained continuously awake for \( \approx 32 \) h in a semirecumbent position in bed with ambient illumination <15 lux (10, 11, 24). The purpose of the constant routine is to eliminate or reduce variables that might otherwise obscure or confound the expression of circadian rhythmicity and thereby allow for a more accurate estimate of circadian phase. Instead of receiving three meals and an evening snack, as was done on the baseline days, subjects received equal hourly aliquots of food and liquid with the same caloric and nutritional intake as was received on the baseline days, adjusted for inactivity (16). During the constant routine, subjects were attended at all times to ensure compliance with the protocol. Using the constant routine, we were able to determine the timing of the minimum of the fitted core body temperature rhythm using a two-harmonic regression analysis (6). The constant routine ended 9.5 h after the calculated time of the fitted minimum of the core body temperature rhythm.

On days 6–8, subjects lived on a shifted sleep-wake schedule, going to sleep at \( \approx 1600 \) and awakening at \( \approx 2400 \) (the center of the 8-h sleep episode was scheduled to be 13.5 h after the time of the fitted minimum of core body temperature as determined during the initial constant routine). Lighting was dim (<15 lux) during the wake period of these 3 days, except for a 5-h experimental light stimulus scheduled to be centered 1.5 h after the estimated position of the minimum of core body temperature as determined during the first constant routine (i.e., midnight was 12 h opposite midsleep). Experimental light stimuli consisted of exposure to 3 consecutive days of 5 h of <0.03 lux (\( n = 8 \)), 12 lux (\( n = 8 \)), 180 lux (\( n = 8 \)), 600 lux (\( n = 9 \)), 1,260 lux (\( n = 8 \)), or 9,500 lux (\( n = 7 \)). Results from the <0.03 and 9,500 lux condition have, in part, been reported by Duffy and colleagues in 1996 (15). Results from the 12-lux condition have, in part, been reported by Zeitzer and colleagues in 1997 (37). Results from the 180- and 1,260-lux condition have, in part, been reported by Boivin and colleagues in 1996 (3). During each stimulus condition, except the 9,500-lux exposure, subjects alternated their gaze from a fixed spot on the wall to a free gaze around the room every 10 min. In the 9,500-lux condition, subjects alternated their gaze from fixed to free every 10 min, but the fixed gaze was not at a single location in the room. Light measurements reported herein refer to the average light exposure during the fixed gaze portion of the light exposure. For the 25 min preceding and following this 5-h period, during which time subjects maintained a fixed gaze throughout, light intensity was in five steps (16%, 32%, 50%, 63%, 80%) incrementally raised or lowered, respectively, from the dim light to the experimental light value in an effort to reproduce the gradual changes in light intensity observed in nature. These increments were created using a step-wise change in the number of lamps or by a series of goggles (Ultraguard 9400 welding goggles; Uvex Winter Optical, Smithfield, RI), with each goggles set with the appropriate Kodak neutral density filter. Upon awakening on day 9 and until bedtime on day 10, subjects underwent a second constant routine of \( \approx 48 \) h to reevaluate the position of the circadian pacemaker. On day 11, following an 8-h sleep episode, subjects were discharged or continued with other protocols.

Data analysis. Melatonin phase has been shown to be a more accurate representation of the timing of the human circadian pacemaker than core body temperature phase (22). We determined the phase of the melatonin rhythm during the first and second constant routines. Phase shift was calculated as the phase during the first constant routine less the phase during the second constant routine. Melatonin phase was calculated as the midpoint between the time at which the plasma melatonin concentration rose past the 24-h average and the time at which the plasma melatonin concentration fell below the 24-h average (22, 28, 35). If necessary, the precise time at which the melatonin concentration rose above or fell below the 24-h average was interpolated from the sample values taken at the two adjacent time points. In addition to the calculation of the phase of melatonin, we also determined the degree of the suppression of plasma melatonin during the first experimental light episode. Suppression was calculated as: (AUCbaseline – AUClight)/AUCbaseline, in which AUC was the area under the curve as calculated by the trapezoidal method (Origin 5.0; Microcal, Northampton, MA). Baseline AUC was evaluated 2–5 h after the melatonin midpoint during the first constant routine, during which time the subject was continuously semirecumbent and exposed to <15 lux of light. Light AUC was calculated during the same 3 clock h of the next melatonin cycle, during which time all subjects were seated and exposed to the experimental light. Analysis was limited to the first day of experimental light as the phase of the melatonin rhythm would be differentially shifted in the different experimental conditions and the magnitude of suppression may be dependent on the phase of application (23). Because of blood collection difficulties, one subject was excluded from both phase-change and suppression analyses (subject 1008.02, exposed to <0.03 lux), two subjects were excluded from phase-change analysis only (subject 1392, exposed to 180 lux; subject 1309, exposed to 1,260 lux), and another seven subjects were excluded from melatonin suppression analysis only (subjects 1010 and 1131, exposed to <0.03 lux; subject 1402, exposed to 180 lux; subject 1749, exposed to 600 lux; subjects 1015, 1137, and 1228, each exposed to 9,500 lux).

Models. The following models (Tables 1 and 2) were fit to the data: log, power, cube root (power model with the power term fixed at 0.33)

<table>
<thead>
<tr>
<th>Model</th>
<th>Formula</th>
<th>Parameter Estimates</th>
<th>Adjusted R²</th>
<th>AIC</th>
<th>RSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Log</td>
<td>( y = a + b \ln x )</td>
<td>( a = -0.650 \pm 0.313 )</td>
<td>( b = 0.526 \pm 0.0521 )</td>
<td>0.69</td>
<td>81.75</td>
</tr>
<tr>
<td>3-Parameter logistic</td>
<td>( y = \frac{a-c}{1+(xb)} + c )</td>
<td>( a = -1.38 \pm 0.355 )</td>
<td>( b = 113 \pm 48.5 )</td>
<td>( c = 4.07 \pm 0.360 )</td>
<td>0.77</td>
</tr>
<tr>
<td>4-Parameter logistic</td>
<td>( y = \frac{a-c}{1+(xb^2)} + c )</td>
<td>( a = -1.55 \pm 0.455 )</td>
<td>( b = 107 \pm 53.8 )</td>
<td>( c = 4.28 \pm 0.553 )</td>
<td>( d = 0.803 \pm 0.257 )</td>
</tr>
<tr>
<td>Power</td>
<td>( y = a \cdot x^b )</td>
<td>( a = 0.402 \pm 0.191 )</td>
<td>( b = 0.272 \pm 0.0581 )</td>
<td>0.58</td>
<td>112.2</td>
</tr>
<tr>
<td>Cube root</td>
<td>( y = a \cdot \sqrt[3]{x} )</td>
<td>( a = 0.242 \pm 0.0226 )</td>
<td>0.57</td>
<td>114.2</td>
<td>2.55</td>
</tr>
</tbody>
</table>

Model fits to phase shifts in response to 3 consecutive episodes of experimental light exposure. See MATERIALS AND METHODS for description of the models and statistics. Parameters are shown with the SD of the fit. AIC, Akaike’s information criterion; RSE, residual square error.
ILLUMINANCE RESPONSE OF THE HUMAN CIRCADIAN SYSTEM

Table 2. Model fits for illuminance-induced suppression of plasma melatonin

<table>
<thead>
<tr>
<th>Model</th>
<th>Formula</th>
<th>Parameter Estimates</th>
<th>Adjusted R²</th>
<th>AIC</th>
<th>RSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Log</td>
<td>( y = a + b \cdot \ln x )</td>
<td>( a = 0.209 \pm 0.0695 )</td>
<td>0.53</td>
<td>5.80</td>
<td>0.0500</td>
</tr>
<tr>
<td>3-Parameter logistic</td>
<td>( y = \frac{a-c}{1+(x/b)^c} + c )</td>
<td>( a = 0.185 \pm 0.0783 ), ( b = 127 \pm 85.0 ), ( c = 0.877 \pm 0.0765 )</td>
<td>0.58</td>
<td>7.57</td>
<td>0.0448</td>
</tr>
<tr>
<td>4-Parameter logistic</td>
<td>( y = \frac{a-c}{1+(x/b)^c} + c )</td>
<td>( a = 0.113 \pm 0.129 ), ( b = 84.2 \pm 76.9 ), ( c = 0.901 \pm 0.132 ), ( d = 0.719 \pm 0.357 )</td>
<td>0.57</td>
<td>9.56</td>
<td>0.0459</td>
</tr>
<tr>
<td>Power</td>
<td>( y = a \cdot x^b )</td>
<td>( a = 0.272 \pm 0.0566 ), ( b = 0.142 \pm 0.0294 )</td>
<td>0.51</td>
<td>5.89</td>
<td>0.0526</td>
</tr>
<tr>
<td>Cube root</td>
<td>( y = a \cdot x^{0.33} )</td>
<td>( a = 0.0618 \pm 0.00532 )</td>
<td>0.046</td>
<td>5.78</td>
<td>0.102</td>
</tr>
</tbody>
</table>

Model fits to acute suppression of plasma melatonin during hours 2–5 after the presumptive midpoint of the peak of melatonin during the 1st experimental light exposure. See MATERIALS AND METHODS for description of the models and statistics. Parameters are shown with the SD of the fit.

(3), three-parameter logistic (from which the Naka-Rushton and Michaelis-Menten equations are derived) (25), and a four-parameter logistic [a version of the three-parameter logistic model with an added power term that has been shown to be useful in modeling some biological responses to light (4, 5, 25)]. In the logistic models, the \( a \) term is the estimated response of the system to 0 lux of light, the \( b \) term is the lux value at which 50% of the maximal response is observed, the \( c \) term is the asymptotic maximal response of the system, and the \( d \) term, in the four-parameter logistic model, is a measure of the steepness of the rising portion of the curve. Data were fit with a nonlinear least-squares fitting analysis based upon the Levenberg-Marquardt method (CurveExpert 1.34; D. Hyams, Starkville, MS; Microcal Origin 5.0, Microcal). The goodness of fit of each model was assessed by calculating the adjusted \( R^2 \), Akaike’s information criterion (AIC) (26), and the residual square error (RSE, sum of the square of the residuals divided by degrees of freedom). When fitting the phase shift data, in both the logistic models, we constrained the \( a \) term from \(-0.96\) to \(-0.24\), as this is the average estimated drift (i.e., movement of the clock unaltered by the environment) between the two phase estimations (9, 15). We also examined the logistic models leaving the \( a \) term unconstrained and found that the goodness-of-fit measures did not significantly change (three-parameter logistic model: adjusted \( R^2 = 0.77 \), AIC = 10.84, RSE = 0.269; four-parameter logistic model: adjusted \( R^2 = 0.76 \), AIC = 12.77, RSE = 0.281; compare with Table 1). When fitting the acute suppressive effects of light on plasma melatonin concentration, we constrained the \( c \) term of the logistic models to be \( \leq 1 \), as the maximal response of the system is 1 (i.e., 100% suppression).

RESULTS

Shifting of the timing of plasma melatonin. Three consecutive days of experimental light during the late biological night caused an intensity-dependent increase in both the phase shift and acute suppression of plasma melatonin. Exposure to 3 consecutive days of experimental light on average (± SE) shifted the midpoint of the peak of the melatonin rhythm by \(-1.53 \pm 0.16 \text{ h} \) (\( n = 7, <0.03 \text{ lux} \)), \(-0.69 \pm 0.30 \text{ h} \) (\( n = 8, 12 \text{ lux} \)), \(1.80 \pm 0.40 \text{ h} \) (\( n = 7, 180 \text{ lux} \)), \(3.75 \pm 0.58 \text{ h} \) (\( n = 9, 600 \text{ lux} \)), \(2.78 \pm 0.42 \text{ h} \) (\( n = 7, 1,260 \text{ lux} \)), and \(4.33 \pm 0.33 \text{ h} \) (\( n = 7, 9,500 \text{ lux} \)) (Fig. 1A). The observed \(-1.5\)-h delay

![Fig. 1. Phase shifts (A) and melatonin suppression (B) in response to 3 consecutive days of experimental light. The data are shown as means ± SE and are fitted with a 3-parameter logistic fit (solid line) with upper and lower 95% confidence intervals (dotted line). The logistic fit was calculated using a nonlinear least-squares analysis (see MATERIALS AND METHODS). See Tables 1 and 2, respectively, for model statistics.](http://ajpregu.physiology.org/)
in the $<0.03$ lux control is likely due to a drift of the circadian pacemaker of $0.3$ h per day for the 5-day interval between phase determination (9). Compared with the $<0.03$ lux control, each of the other experimental light exposures induced a significant shift of the midpoint of the peak of the melatonin rhythm $P < 0.05$ (12 lux), $P < 0.01$ (180, 600, 1,260, 9,500 lux); one-tailed $t$-tests. Saturating phase shifts appeared to occur after exposure to illuminances $>1,000$ lux.

Acute suppression of plasma melatonin. Acute suppression of plasma melatonin during the first of the three experimental light exposures also exhibited an intensity-dependent relationship, with higher illuminances inducing greater suppression. In general, illuminance $<15$ lux induced little suppression, whereas illuminances $>200$ lux induced complete suppression. The group of subjects exposed to 180 lux exhibited varying degrees of suppression ranging from complete to minimal. In response to the experimental light exposure, the plasma melatonin concentration of subjects, on average ($\pm$SE), acutely suppressed (i.e., reduced) $9.74 \pm 11.1\%$ ($n = 3$, $<0.03$ lux), $29.8 \pm 10.2\%$ ($n = 8$, 12 lux), $52.3 \pm 12.4\%$ ($n = 7$, 180 lux), $82.2 \pm 4.65\%$ ($n = 8$, 600 lux), $82.0 \pm 2.64\%$ ($n = 8$, 1,260 lux), and $81.1 \pm 2.04\%$ ($n = 4$, 9,500 lux) (Fig. 1B). Comparison of the experimental light exposure to the $<0.03$ lux control group was not statistically possible as, because of a lack of blood sampling in most of these subjects during their first experimental exposure to $<0.03$ lux, acute suppression of melatonin could only be calculated in three subjects. However, compared with the 12-lux group, groups exposed to an illuminance of 600 lux or higher significantly suppressed plasma melatonin ($P < 0.01$, one-tailed $t$-tests). The group exposed to 180 lux exhibited acute melatonin suppression that was not quite significantly different from the group exposed to 12 lux ($P = 0.09$, one-tailed $t$-test).

Modeling phase shift data. To quantify the response of the human circadian timing system to light during the late subjective night, we fit the phase shift data with the following models (see MATERIALS AND METHODS and Table 1): log, power, cube root, three-parameter logistic, and four-parameter logistic. Each of the models significantly fit the phase shift data ($P < 0.01$, approximate $F$-tests), but the logistic models best described these data. Both logistic models estimate a maximal response of the human circadian timing system to 3 consecutive days of light exposure of this duration and timing to be $\sim 4$ h, with a half-maximal response occurring at $50–160$ lux, a range typical of ambient, indoor illumination (Table 1, Fig. 1A). The goodness-of-fit measures and the variance of the parameter estimates indicate that the three-parameter model is slightly less variable in estimating the model parameters than is the four-parameter logistic model (Table 1).

Modeling suppression data. As with the phase shift data, we also fit the melatonin suppression data with the log, power, cube root, three-parameter logistic, and four-parameter logistic models (Table 2). Each model significantly fit the data ($P < 0.01$, approximate $F$-tests), though the logistic models fit the data best. The logistic models estimate a maximum suppression to light administered at this phase to be $\sim 90\%$ (Table 2, Fig. 1B). The logistic models also estimate that the illuminance at which $50\%$ of the maximum suppression occurs is in the room light range; however, there is substantial variance associated with estimation of this parameter (Table 2).

DISCUSSION

Our data indicate that the human circadian pacemaker responds to increasing illuminance administered at a noncritical phase during the late subjective night in a monotonically increasing manner. Rather than linearly rising to a log, power, or cube-root compression of illuminance, the human circadian system appears to follow logistic dynamics in response to light delivered during the late biological night, as has been demonstrated for light administered during the early biological night (36). Logistic dynamics have also been used to describe the photic dose response of the circadian timing system of nonhuman mammals (1, 5, 25, 29, 32). In response to 3 consecutive days of 5 h of experimental light exposure, the pacemaker has a half-maximum response that is achieved with illuminances of $\sim 50–160$ lux (typical of indoor ambient illumination) and saturates at $\sim 1,400$ lux.

Acute suppression of plasma melatonin by light was also illuminance dependent. Model fitting indicates that logistic kinetics best explain the relationship between illuminance and acute melatonin suppression. As with the light-induced phase shifts of the melatonin rhythm, the logistic model fit showed that half of the maximal suppression of melatonin occurred in the room light range, $10–200$ lux, and saturated at $\sim 1,100$ lux. A large standard deviation is associated with calculation of the half-maximum term ($b$), as there is little data for response to illuminances in the portion of the fitted curve that has the greatest derivative (largest slope).

Even though these phase advance shift data were garnered using a 5-h pulse of experimental light exposure, with additional 25-min transition periods before and after) administered on 3 consecutive days during the phase advance portion of the phase response curve (i.e., late biological night), they indicate a similar photic sensitivity of the human circadian pacemaker as did a less complex, single episode of $6.5$ h of experimental light exposure (no transitions before or after) on a single day in the phase delay region of the phase response curve (i.e., early biological night) (36) (Fig. 2). Both data sets indicate that...
ordinary indoor room light is capable of significantly resetting the human circadian pacemaker and that such light is \( \approx 50\% \) as effective in resetting the human circadian pacemaker as is light nearly 100-fold brighter. As can be observed in Fig. 2, based on logistic model fits, the single pulse response appears to cause a saturating response at \( \approx 550 \) lux, whereas the pulse that consists of 3 days of experimental light exposure appears to cause a saturating response at \( \approx 1,400 \) lux. This, however, may be due to the nonlinear phase-dependent effects of light on the human circadian pacemaker that occur in the three-exposure experiment. Both this current experiment and a previous experiment (36) examined the dose-response relationship between the circadian timing system and light at times during which the phase-shifting responsiveness of the clock resembles simple, phase-only resetting (18). It will, therefore, be important to examine this dose-response relationship near the cross-over point of the phase-response curve (i.e., in the critical region) where either very small or very large phase changes occur and both phase and amplitude of the circadian oscillation are reset by light (11).

Although we have shown that the human circadian pacemaker is exquisitely sensitive to light, such that even 10 lux of light is capable of inducing significant changes in the timing of the pacemaker, the question still remains as to the functional significance of these findings. The subjects in this study were exposed to \( >48 \) h of light of \( <10 \) lux intensity before receiving the first experimental light exposure. It is possible that this “background” light may significantly influence the subsequent responsiveness of the circadian timing system (17, 30). Our data, therefore, cannot be taken to imply that normal room light is always half as potent as outdoor light, just that it has the capacity to be as such and cannot be ignored in the context of shift work or jet travel. Another aspect of our data to consider is the differential contribution of the two ocular photoreceptive systems (three cones vs. intrinsically photosensitive retinal ganglion cells) to the circadian dose-response curve to light. As these two systems likely have overlapping ranges of intensity at which they are active yet have distinct spectral sensitivities, how spectral efficiency changes with changes in intensity remains to be determined.

ACKNOWLEDGMENTS

We thank Dr. D.-I. Dijk for helpful comments and advice. Current addresses: J. M. Zeitzer, Stanford Center for Narcolepsy Research, Stanford University Medical Center, Palo Alto, CA 94305; D. B. Boivin, Douglas Hospital Research Centre, McGill Medical School, Verdun, QC, Canada.

GRANTS

S. B. S. Khalsa was supported by a senior National Research Service Award fellowship from the National Heart, Lung, and Blood Institute. This research was supported by the National Aeronautics and Space Administration, National Institute of Mental Health, National Institutes of Health, National Space Biomedical Research Institute, and the General Clinical Research Center Program of the National Center for Research Resources.

REFERENCES


