Original article

# Reciprocal Relations Between Objectively Measured Sleep Patterns and Diurnal Cortisol Rhythms in Late Adolescence 

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## A B S T R A C T

Purpose: To examine how hours of sleep and wake times relate to between-person differences and day-today changes in diurnal cortisol rhythms in late adolescence.
Methods: Older adolescents $(\mathrm{N}=119)$ provided six cortisol samples (wakeup, +30 minutes, +2 hours, +8 hours, +12 hours, and bedtime) on each of three consecutive days while wearing an actigraph. We examined how average (across 3 days) and day-to-day changes in hours of sleep and wake times related to diurnal cortisol patterns.
Results: On average, more hours of sleep related to steeper decline in cortisol across the days. Day-to-day analyses revealed that the hours of sleep of the previous night predicted steeper diurnal slopes the next day, whereas greater waking cortisol levels and steeper slopes predicted more hours of sleep and a later wake time the next day.
Conclusion: Our results suggest a bidirectional relationship between sleep and hypothalamic-pituitaryadrenal axis activity.
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Adolescence is marked by a biological shift in sleep patterns often resulting in more "owl-like" patterns of sleep (e.g., later bed times, later wake times) [1-3]. Such changes often conflict, particularly in later adolescence, with the growing social demands that many individuals face $[2,4]$. Findings indicate that late adolescents report more erratic sleep schedules, later waking times, and less sleep as compared with early adolescents [4]. Less than $30 \%$ of late adolescents get the recommended $\geq 8$ hours of sleep per night, and nearly $40 \%$ of individuals report consistent poor sleep quality $[5,6]$. Such sleep patterns have negative implications for individuals' outcomes, including mood disorders [7], difficulties in school [8,9], and physical health (e.g., obesity [10]).

Sleep patterns have also been known to play an important role in regulating other aspects of physiology, including the hy-

[^0]pothalamic-pituitary-adrenal (HPA) axis. The main hormone of HPA axis (cortisol) exhibits a strong diurnal rhythm, with high levels at wakeup time, increasing further to a peak 30 minutes after waking (called the cortisol awakening response [CAR]) and then decreases rapidly across the day, reaching nadir around midnight [11]. These changes in cortisol coincide with individuals' sleep cycles; low levels of cortisol are present during the first half of the night which is dominated by deep sleep (or slow wave sleep), while high levels of cortisol are present during the second half of the night which is dominated by rapid eye movement (REM) sleep (more wakeful period) [12,13].

This correspondence has been the topic of research, primarily among adults, showing a bidirectional and complex relation between sleep and HPA axis functioning. The HPA axis functioning has been shown to influence sleep patterns [14,15], and changes in sleep patterns, specifically wake times and hours of sleep, have been found to influence daytime cortisol rhythms $[16,17]$. Surprisingly, however, the relation between sleep and HPA axis functioning has yet to be examined among adolescents. This is an important gap, given that adolescence is a critical developmental

Table 1
Descriptive statistics for study variables $(\mathrm{N}=119)$

| Variable | \% | n | Mean | SD | Range |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Age of adolescent |  |  | 19.01 | . 42 | 17.93-20.10 |
| Wake time (self-report) hh:mm |  |  | 08:38 | 1:28 | 03:13-13:30 |
| Wake time (Actigraph) hh:mm |  |  | 08:47 | 1:22 | 05:48-12:33 |
| Hours of night-time sleep (self-report) |  |  | 7.71 | 1.34 | 3.00-10.61 |
| Hours of night-time sleep (actigraph) |  |  | 6.41 | 1.07 | 1.41-9.55 |
| Sleep efficiency \% (actigraph) |  |  | 81.85 | 7.61 | 48.80-92.60 |
| Hours of naptime (self-report) |  |  | . 41 | . 77 | .00-04.72 |
| Average wake time cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 30 | . 21 | .01-1.37 |
| Average +30 minute cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 50 | . 25 | .01-1.78 |
| Average +3 hours cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 28 | . 20 | .03-1.80 |
| Average +8 hours cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 16 | . 11 | .03-. 82 |
| Average +12 hours cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 12 | . 11 | .01-.76 |
| Average bedtime cortisol ( $\mu \mathrm{g} / \mathrm{dL}$ ) |  |  | . 10 | . 11 | .01-. 71 |
| Caffeine (drinks per day) |  |  | . 91 | 1.40 | .00-8.00 |
| Nicotine (cigarettes per day) |  |  | . 28 | . 82 | .00-5.00 |
| Alcohol (drinks per week) |  |  | 4.38 | 7.45 | .00-50.00 |
| Exercise (hrs/wk) |  |  | 2.77 | 4.07 | .00-30.00 |
| Oral contraceptive use (females only) | 35.5 | 33 |  |  |  |
| Asthma medication | 7.6 | 9 |  |  |  |
| Depression medication | 4.2 | 5 |  |  |  |
| Male | 21.8 | 26 |  |  |  |
| European American | 61.4 | 73 |  |  |  |
| African American | 17.6 | 21 |  |  |  |
| Hispanic | 4.2 | 5 |  |  |  |
| Multiple/other race | 16.8 | 20 |  |  |  |

period in which physiological changes occur $[18,19]$ and sleep patterns are shifting (especially among late adolescents [20]). Further, recent research has linked diurnal cortisol functioning, specifically, flattened diurnal slopes across the waking day, to a variety of physical and mental health outcomes [21-24]. A better understanding of the relation of nighttime sleep and daytime cortisol rhythms may have important implications for our understanding of the development of a variety of disorders related to both sleep and the HPA axis. In this study, we take an initial step in trying to understand the associations between adolescent sleep and diurnal cortisol by examining how hours of sleep and wake times relate to adolescents' waking cortisol values, CAR, and diurnal decline across the waking day. Utilizing multiple salivary samples over consecutive days, we first examined betweenperson differences by investigating how individuals' average (across days of sampling) hours of sleep and average wake times related to average cortisol rhythms. Then, to gain a better understanding of the reciprocal relation between sleep and HPA axis functioning, we investigated how within-person day-to-day variations in sleep patterns (i.e., wake time, hours of sleep) related to day-to-day variations in diurnal cortisol.

## Methods

## Participants

Data for the current study were obtained from a larger longitudinal study that focused on adolescents from two diverse public high schools (one in the Midwest, and the other on the West Coast). For the larger study, participants with high neuroticism levels [25] were oversampled, resulting in $61 \%$ of the sample scoring in the top third of the neuroticism screener. A total of $79 \%$ of the Midwest participants ( $\mathrm{n}=243$ ) were randomly invited to participate in a longitudinal cortisol sampling protocol. Of the adolescents invited, 173 (71\%) agreed to participate and completed wave 1 cortisol sampling. The current study uses data
from 152 adolescents who participated in wave 2 of cortisol collection (approximately 1.5 years after wave 1 ), at which point actigraph measures of sleep were added. Adolescents who were pregnant ( $\mathrm{n}=1$ ), currently taking steroids ( $\mathrm{n}=7$ ), or had missing data on study variables $(\mathrm{n}=25)$ were excluded. Thus, 119 adolescents ( $77 \%$ female) participated in the study. The greater proportion of females is accounted for by the fact that on an average they report higher levels of neuroticism [26]. Of the current study's sample, $64.7 \%$ scored in the top third of the neuroticism screener. Differences on all study variables by neuroticism risk score were examined and revealed that high risk individuals reported greater caffeine use compared to low- or medium-risk individuals $[F(2,116)=3.04, p=.05]$, and low-risk individuals were more likely to be taking asthma medication than the other two groups $\left[\chi^{2}(2)=9.02, p<.05\right]$. Adolescents ranged in age from 17.93 to 20.10 years ( $M=19.01$ ) and were from varying ethnic and/or racial backgrounds (Table 1).

## Procedures

Participants were sent a study packet that contained an actigraph [27], diary booklets, a mechanical kitchen timer (to assist with the timing of second sample), straws, vials, labels, and a health and medical questionnaire. Study personnel contacted participants to review the study protocol. The participants provided six salivary cortisol samples and completed six diary entries per day for three consecutive typical weekdays. Participants wore an actigraph the night before starting the cortisol sampling and left it on until the morning after the last day of the study. Participants were paid $\$ 30$ for completion of the sampling protocol and given a summary of basic sleep statistics across the 3 days of sampling. As approved by our Institutional Review Board, individual sleep data were used only for descriptive and hypothesis testing purposes, not for assessment or diagnosis of sleep disorders.

## Measures

## Salivary cortisol

Saliva samples were gathered each day for 3 days: at wakeup, 30 minutes after waking, at bedtime, and three semirandom times throughout the day signaled by the actigraph watch (approximately 2,8 , and 12 hours post-awakening). Participants were asked for their typical wake time for each day of the study and watches were programmed to signal 2,8 , and 12 hours from this time (day 2). Thirty-minute variations were added for days 1 and 3 to make signals less expected (i.e., day 1 participants took samples 30 minutes early [1.5, 7.5, and 11.5 hours from waking] and on day 3 they took the samples 30 minutes later [2.5, 8.5 , and 12.5 hours from waking]). Participants expelled saliva through a small straw into a 2 mL polypropylene tube and labeled tubes with the time and date. Participants were instructed not to eat, drink or brush their teeth 30 minutes before sampling. Samples were returned by mail, refrigerated at $-20^{\circ} \mathrm{C}$, and then sent on dry ice by courier to Biochemisches Labor, Trier, Germany, to be assayed for cortisol. Cortisol levels are stable at room temperature for several weeks and are unaffected by shipping [28]. Assays were conducted using a time-resolved immunoassay with fluorometric detection (see reference [29] for greater assay description). Intra-assay coefficients of variation were between $4.0 \%$ and $6.7 \%$, and inter-assay coefficients of variation ranged from $7.1 \%$ to $9.0 \%$.

## Objective sleep

During the 3-day salivary cortisol data collection, individuals wore the Actiwatch Score (Phillips Respironics, Inc.), a wristbased accelerometer placed on the nondominant hand that quantifies movement across the waking day and during sleep. To score data, the Actiware-Sleep software (version 3.4, MiniMitter/ Philips Respironics)-validated algorithm was used [30]. Utilizing 1-minute epochs and based on significant movement after at least 10 minutes of inactivity, this algorithm calculates a variety of sleep parameters including sleep end (wake time) and hours of sleep (sleep time excluding all periods of wakefulness during the total sleep period) [31]. Actigraph sleep estimates have been validated against concurrent polysomnography [32]. Aggregate sleep parameters were created by averaging individuals' sleep parameters across the three sampling days; sleep parameters for each day were used in the day-to-day analyses.

## Diary and health variables

Using paper and pencil diaries, adolescents reported on sleep and health behaviors for each day of cortisol sampling. Specifically, adolescents reported their waking time and previous night's bedtime, as well as any duration of time spent napping during the day. Adolescents also completed diary entries with each saliva sample, reporting whether they consumed caffeinated drinks, cigarettes, alcohol, medicines, or exercised within an hour of each sample. In a health questionnaire, youth reported whether they were taking oral contraceptives (females only) or anti-depressant medication.

## Analytic plan

A three-level multilevel growth-curve analysis was used to account for the nested nature of our data $[33,34]$. This modeling controls nonindependence associated with nesting and allows for levels of cortisol to be predicted by moment-level variables

Table 2
HLM 3-level equations modeling diurnal cortisol

$$
\begin{aligned}
& \text { Equation } 1 \\
& \text { Level } 1 \\
& \quad \text { Cortisol } \\
& \text { Level } 2 \\
& \pi_{0 i j}=\gamma_{00 j}+\zeta_{0 i j} \\
& \pi_{1 i j}=\gamma_{10 j}+\zeta_{1 i} \\
& \pi_{2 i j}=\gamma_{20 j}+\zeta_{2 i} \\
& \text { Level } 3 \\
& \quad \gamma_{00 j}=\beta_{000}+\beta_{001} \text { (aggregate sleep parameter) }+r_{00 j} \\
& \gamma_{10 j}=\beta_{100}+\beta_{101} \text { (aggregate sleep parameter) }+r_{10 j} \\
& \gamma_{20 j}=\beta_{200}+\beta_{201}(\text { aggregate sleep parameter })+r_{20 j} \\
& \text { Equation } 2 \\
& \text { Level } 2 \\
& \pi_{0 i j}=\gamma_{00 j}+\gamma_{01 j} \text { (sleep parameter before Cort) }+\gamma_{02 j} \text { (sleep parameter } \\
& \text { after Cort) }+\zeta_{0 i} \\
& \pi_{1 i j}=\gamma_{10 j}+\gamma_{11 j} \text { (sleep parameter before Cort) }+\gamma_{12 j} \text { (sleep parameter } \\
& \text { after Cort) }+\zeta_{1 i j} \\
& \pi_{2 i j}=\gamma_{20 j}+\gamma_{21 j} \text { (sleep parameter before Cort) }+\gamma_{22 j} \text { (sleep parameter } \\
& \text { after Cort) }+\zeta_{2 i j}
\end{aligned}
$$

(level 1), day-varying variables (level 2), and individual-level variables (level 3). In line with previous studies [34,35], day-level variables were added, both lagged (minus 1 day) and nonlagged (night after cortisol collection) at level 2 to simultaneously model the effect of sleep parameters of earlier night on next-day cortisol, and the effect of cortisol on sleep later that evening. As recommended [36], level 2 variables were centered within cluster and level 3 variables were centered at the grand mean. At level 1, time was centered as hours since waking (e.g., waking $=0$ ).

We first modeled the latent estimates of the parameters defining each individual's diurnal cortisol rhythm. Next, we entered individuals' 3-day average sleep parameters (i.e., wake time, hours of sleep) at level 3 to examine their associations with average cortisol levels (Equation 1, Table 2). Cortisol values are predicted by the time of each sample, scaled as hours since waking each day, such that the $\beta_{000}$ (the intercept) reflects the average wakeup cortisol level across individuals, $\beta_{100}$ reflects individuals' average CAR (Individuals' second sample [30 minutes after waking] was dummy coded, in which $1=$ second cortisol sample and $0=$ all other samples) and $\beta_{200}$ reflects the average linear slope of participants' diurnal cortisol rhythms. The current analysis did not include a quadratic time variable because the inclusion of this term in the unconditional growth model did not improve our model fit $\left[\chi^{2} \Delta(1)=.78, p=.37\right]$. Given this, the slope can be interpreted as a linear decline across the day rather than the linear decline at waking. Coefficients $\beta_{001}, \beta_{101}$, and $\beta_{201}$ reflect the effect of average sleep parameters on average wakeup cortisol level, average CAR, and average linear slope, respectively. Average wake time and sleep were entered in separate models, followed by an analysis that included both.

Finally, to understand the effect of changes in sleep parameters on day-to-day changes in cortisol rhythms within individuals, we examined the relation of sleep parameters the night before and after cortisol sampling to diurnal cortisol profiles each day by entering day-level sleep parameters at level 2 (Equation 2, Table 2). For wake time, the sleep parameter before cortisol refers to the wake time in the morning before cortisol sampling that day, whereas sleep parameter after cortisol refers to wake time the morning after cortisol sampling. For hours of sleep, the sleep parameter before cortisol refers to the previous night's hours of sleep before the start of the next day's cortisol sampling, whereas the sleep parameter after cor-

Table 3
Effects of average wake time and hours of sleep on diurnal cortisol

| Fixed effects | $\begin{gathered} \text { Model 1 } \\ \text { Coefficient (SE) } \end{gathered}$ | $\begin{gathered} \text { Model } 2 \\ \text { Coefficient (SE) } \end{gathered}$ | $\begin{gathered} \text { Model 3 } \\ \text { Coefficient (SE) } \end{gathered}$ |
| :---: | :---: | :---: | :---: |
| Wake up cortisol level |  |  |  |
| Intercept | $-1.423(.059)^{* * *}$ | -1.423 (.059)*** | $-1.424(.060)^{* * *}$ |
| Average wake time | -. 005 (.036) |  | -. 009 (.036) |
| Average hours of sleep |  | . 032 (.051) | . 036 (.052) |
| CAR |  |  |  |
| Intercept | . 544 (.041)*** | . $543(.042)^{* * *}$ | . 544 (.041)*** |
| Average wake time | . 017 (.032) |  | . 020 (.032) |
| Average hours of sleep |  | -. 014 (.043) | -. 019 (.042) |
| Time since waking |  |  |  |
| intercept | $-.091(.005)^{* * *}$ | -. 092 (.005)*** | -. 091 (.005**** |
| Average wake time | . 008 (.004) |  | . 010 (.004)* |
| Average hours of sleep |  | -. 009 (.005)* | -. 012 (.005)* |

Level 1 ( $n=1521$ ), level $2(n=285)$, level $3(n=119)$
Note: ${ }^{*} p \leq .05,{ }^{* * *} p<.001$. Race/ethnicity, gender, birth control, asthma medication, depression medication, and daytime sleep (naps) were added as covariates at level 3. Caffeine use, nicotine use, and alcohol use, any medication and exercise were added as covariates at level 1.
tisol refers to the hours of sleep after that day's cortisol sampling. Coefficients $\gamma_{01 j}$, $\gamma_{11 j}$, and $\gamma_{21 j}$ reflect the relation of the sleep parameter the night before cortisol collection to subsequent day's wakeup cortisol level, CAR, and linear slope, respectively, whereas coefficients $\gamma_{02 j}, \gamma_{12 j}$, and $\gamma_{22 j}$ reflect the relation of wakeup cortisol, CAR, and linear slope, respectively, to the sleep parameters taken that night, after cortisol sampling each day. Wake time and average hours of sleep were entered first separately and then simultaneously in the final model. For all analyses, adolescents' diary reports of their caffeine, nicotine, and alcohol consumption, exercise, and medication use within the hour before each sample were entered as covariates at level 1, whereas birth control, asthma, depression medication, and daytime sleep (naps) were entered as covariates at level 3.

## Results

Cortisol values are presented in original micrograms/deciliter units in Table 1; natural log transformed values were used in all analyses because cortisol values were skewed (2.48) and kurtotic (9.57). In line with recommendations [37], cortisol values were top coded at $1.80 \mu \mathrm{~g} / \mathrm{dL}$ (equivalent to $50 \mathrm{nmol} / \mathrm{L}$ ). The base model showed the expected diurnal patterns; high values on awakening ( $\beta_{000}=-1.40$, standard error (SE) $=.06, p<.0001$; equivalent to $.25 \mu \mathrm{~g} / \mathrm{dL}$.), a strong increase ( $68 \%$ ) in levels in the first 30 minutes (CAR; $\beta_{100}=.52, \mathrm{SE}=.04, p<.0001$ ), and approximately an $8 \%$ per hour decline in cortisol levels from wake up to bedtime ( $\beta_{200}=-.09, \mathrm{SE}=.01, p<.0001$ ).

## Between-individual analysis

As noted in Table 3, average wake time did not relate to average diurnal cortisol profiles (model 1). Average hours of sleep (model 2), however, related to the rate of decline in cortisol; greater hours of sleep related to a steeper decline in cortisol across the day ( $1.1 \%$ steeper slope for every additional hour of sleep). These effects were maintained even after controlling for wake time (model 3).

## Within-individual analysis

As noted in Table 4 (model 1), when relating changes in sleep to changes in cortisol rhythms within individuals across
multiple days, same-day wake time was a significant predictor of subsequent wakeup cortisol values, CAR, and the decline in cortisol. That is, adolescents who woke up later that day had higher wakeup cortisol as compared with the days when they woke up earlier ( $9.5 \%$ increase in morning cortisol for every 1 hour later wake time), had a less pronounced CAR (10.5\% decrease in CAR for every 1 hour increase in wake time), and had a steeper decline in cortisol ( $1.3 \%$ steeper slopes for every 1 hour increase in wake time) across the day. In model 2, prior night's hours of sleep predicted wakeup cortisol, CAR, and decline in cortisol. That is, more hours of sleep the previous night predicted higher wakeup cortisol (15.5\% increase for every hour increase in sleep), a lower CAR ( $10.4 \%$ decrease for every hour increase in sleep), and steeper decline in cortisol ( $1.3 \%$ steeper slope for every hour increase in sleep) the next day. Further, greater levels of wakeup cortisol predicted greater hours of sleep the next day.

The analysis examining wake time and hours of sleep together (model 3) revealed that hours of sleep the previous night and the subsequent day both predicted wakeup cortisol levels. That is, more hours of sleep the previous night predicted greater wakeup cortisol the next day, and greater wakeup cortisol that day related to more hours of sleep that night (controlling for hours of sleep the previous night). For diurnal slopes, a similar bidirectional relation emerged. Specifically, previous and next day hours of sleep both predicted steeper decline in cortisol slopes. Thus, more hours of sleep the previous night predicted a steeper decline in cortisol the subsequent day, and steeper slopes that day related to more hours of sleep that night. Finally, next day wake time related to diurnal slopes, suggesting that flatter diurnal slopes that day predicted later waking times the following day.

## Discussion

The present study took an important first step in understanding how typical hours of sleep and wake times relate to cortisol diurnal rhythms and how late adolescents' day-to-day variation in sleep relates to day-to-day variation in cortisol. Our findings suggest that across individuals, average hours of sleep are related to a steeper diurnal decline in cortisol across the day, even after accounting for adolescents' average wake times. These results are consistent with findings among adults [16], suggesting that individuals who sleep more have diurnal cortisol rhythms char-

Table 4
Effects of same day wake time and prior day hours of sleep on diurnal cortisol

| Fixed effects | Model 1 Coefficient (SE) | Model 2 Coefficient (SE) | Model 3 Coefficient (SE) |
| :---: | :---: | :---: | :---: |
| Wake up cortisol level |  |  |  |
| Intercept | $-1.421(.058)^{* * *}$ | $-1.422(.060)^{* * *}$ | -1.418 (.058)*** |
| Same day wake time | . 091 (.036)* |  | -. 002 (.037) |
| Next day wake time | . 007 (.031) |  | -. 055 (.031) |
| Prior day hours of sleep |  | . 144 (.035)*** | . 150 (.040)*** |
| Next day hours of sleep |  | . 079 (.028)** | . 110 (.028)*** |
| CAR |  |  |  |
| Intercept | . 540 (.041)*** | . 544 (.041)*** | . 540 (.041)*** |
| Same day wake time | -. 114 (.054)* |  | -. 056 (.059) |
| Next day wake time | -. 055 (.042) |  | -. 058 (.047) |
| Prior day hours of sleep |  | -. 110 (.043)* | -. 075 (.045) |
| Next day hours of sleep |  | -. 039 (.044) | . 001 (.048) |
| Time since waking |  |  |  |
| Intercept | -. 092 (.005)*** | -. 092 (.005)*** | -. 092 (.005)*** |
| Same day wake time | -. 013 (.004)** |  | -. 009 (.005) |
| Next day wake time | . 007 (.004) |  | . 015 (.004)*** |
| Prior day hours of sleep |  | -. 013 (.004)** | -. 010 (.004)* |
| Next day hours of sleep |  | -. 005 (.004) | -. 015 (.004)** |

Level 1 ( $n=1,521$ ), level $2(n=285)$, level $3(n=119)$.
Note: ${ }^{*} p<.05,{ }^{* *} p<.01,{ }^{* * *} p<.001$. Race/ethnicity, gender, birth control, asthma medication, depression medication were added as covariates at level 3 ; caffeine use, nicotine use, and alcohol use, any medication and exercise were added as covariates at level 1.
acterized by a steeper decline across the day and align with health findings suggesting that such patterns are considered normative and healthy [38]. The potentially bidirectional interrelations between sleep and the HPA axis [12], however, make it difficult to know whether hours of sleep affect changes in diurnal rhythms or whether diurnal rhythms affect hours of sleep.

A better understanding of causal directionality is gained from our day-to-day within-person examination. Findings suggest that on nights when adolescents slept more, they woke up the next morning with higher waking cortisol levels and exhibited a steeper decline in their cortisol across the day. Further, on days adolescents had higher wakeup cortisol values and a steeper diurnal decline in their cortisol, they experienced greater hours of sleep that night and tended to wake up earlier the following day. An alternate way of viewing these day-to-day changes is that on days adolescents' slept less, they woke up the following morning with lower waking cortisol levels and had flatter cortisol slopes that day. On days that adolescents had lower wakeup values and flatter slopes, they subsequently experienced fewer hours of sleep that night and woke up later the next day. It is important to emphasize that these day-to-day analyses examine within-person changes in sleep patterns and diurnal cortisol from one day to the next, rather than trait or between-person differences. As such, these analyses are less subjected to third variable explanations, such as the possibility that stable genetic or personality factors determine both sleep timing and cortisol patterns.

Together, our findings suggest a bidirectional relation between sleep and the HPA axis. While the exact mechanisms underlying the relations between cortisol and the HPA axis are not fully known, our findings could be linked with the correspondence of sleep cycles and cortisol secretion. Deeper sleep accompanies the quiescent period of the HPA axis, whereas greater cortisol output has been found during the later stages of sleep (dominated by REM sleep) [12,13]. Individuals who sleep less and wake up earlier might have less time to "ramp up" their cortisol levels (which happens during REM sleep) and in turn end up with lower awakening cortisol and flatter slopes than individuals who slept more. Further, guided by previous laboratory
findings that suggest that higher levels of cortisol have the ability to suppress slow wave sleep leading to subsequent sleep deprivation [39,40], individuals with flatter slopes, might have a difficult time falling asleep or staying asleep, leading to fewer total hours of sleep. In an attempt to recover from sleep loss during the night, individuals then sleep later the next morning. Given that the specific sleep stages cannot be examined using actigraphy and we do not have overnight cortisol samples, these explanations remain speculative.

In summary, our study is the first to examine these relations in adolescents using an objective sleep measure and examining within-individual covariation between sleep and cortisol in a naturalistic setting. Further, our study examines these relations during a sensitive and unique developmental period in which changes in physiology and sleep patterns are especially relevant [19,20]. Our findings suggest that the choices made about sleep during this time may have consequences for physiology, and in turn, physiology (the HPA axis) attempts to adapt to adolescents' changing daily schedules. Despite these contributions, however, it is important to note several limitations of our study. First, while our findings suggest that sleep relates to fluctuation in diurnal cortisol, the changes were relatively small ( $1 \%-2 \%$ change for diurnal slopes and $9 \%-16 \%$ changes in waking cortisol). Second, we examined a limited number of days in our examination of day-to-day changes and might not be fully capturing the complexities of sleep and its relations to HPA axis functioning over a longer period. Third, we did not use electronic monitoring devices to track the exact timing of cortisol sampling; we relied on participants' self-report of sampling timings. Failure to time samples appropriately, however, would most likely lead to a reduction in the size of our effects, rather than systematic bias in our data. Finally, individuals with greater neuroticism and females were overrepresented in our sample. We found no differences in study variables by neuroticism risk; however, future research should examine whether associations between sleep and HPA axis functioning are similar across personality characteristics and gender. Despite these limitations, our study provides an important first step in understanding the
complex and bidirectional relations between adolescent sleep and HPA axis functioning.

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## References

[1] Carskadon MA, Vieira C, Acebo C. Association between puberty and delayed phase preference. Sleep 1993;16:258-62.
[2] Dahl RE, Lewin DS. Pathways to adolescent health: Sleep regulation and behavior. J Adolesc Health 2002;31(6S):175-84.
[3] Wolfson AR, Carskadon MA. Sleep schedules and daytime functioning in adolescents. Child Dev 1998;69:875-87.
[4] Ohayon MM, Roberts RE, Zulley J, et al. Prevalence and patterns of problematic sleep among older adolescents. J Am Acad Child Adolesc Psychiatry 2000;39:1549-56.
[5] Lund HG, Reider BD, Whiting AB, Prichard JR. Sleep patterns and predictors of disturbed sleep in a large population of college students. J Adolesc Health 2010;46:124-32.
[6] Tsai L, Li S. Sleep patterns in college students: Gender differences and grade differences. J Psychosom Res 2004;56:231-7.
[7] Peterson MJ, Benca RM. Sleep in mood disorders. Sleep Med Clin 2008;3: 231-49.
[8] Fuligni AJ, Hardway C. Daily variations in adolescents' sleep, activities, and psychological well being. J Res Adolesc 2006;16:353-78.
[9] Dewald JF, Meijar AM, Oort FJ, et al. The influence of sleep quality, sleep duration and sleepiness on school performance in children and adolescents: A meta-analysis review. Sleep Med Rev 2010;14:179-89.
[10] Agras WS, Hammer LD, McNicholas F, Kraemer HC. Risk factors for childhood overweight: A prospective study from birth to 9.5 years. J Pediatr 2004;145:20-5.
[11] Pruessner JC, Wolf OT, Hellhammer DH, et al. Free cortisol levels after awakening: A reliable biological marker for the assessment of adrenocortical activity. Life Sci 1997;61:2539-49.
[12] Buckley TM, Schatzberg AF. Review: On the interactions of the hypothalam-ic-pituitary-adrenal (HPA) axis and sleep: Normal HPA axis activity and circadian rhythm, exemplary sleep disorders. J Endocrinol Metab 2005;90: 3106-14.
[13] Born J, Muth S, Fehm HL. The significance of sleep onset and slow wave sleep for nocturnal release of growth hormone (gh) and cortisol. Psychoneuroendocrinology 1988;13:233-43.
[14] Gillian JC, Jacobs LS, Fram DH, et al. Acute effect of glucocorticoid on normal human sleep. Nature 1972;237:398-9.
[15] Bierwolf C, Struve K, Marshal L, et al. Slow wave sleep drives inhibition of pituitary-adrenal secretion in humans. J Neuroendorcinol 1997;9:479-84.
[16] Kumari M, Badrick E, Ferrie J, et al. Self reported sleep duration and sleep disturbance are independently associated with cortisol secretion in the Whitehall II study. J Clin Endocrinol Metab 2009;94:4801-9.
[17] Kudielka BM, Kirschbaum C. Awakening cortisol responses are influenced by health status and awakening time but not by menstrual cycle phase. Psychoneuroendocrinology 2003;28:35-47.
[18] Walker EF, Walder DJ, Reynolds F. Developmental changes in cortisol secretion in normal and at-risk youth. Dev Psychopathol 2001;13:721-32.
[19] Stroud LR, Foster E, Papandonatos GD, et al. Stress response and the adolescent transition: Performance versus peer rejection stressors. Dev Pyschopathol 2009;21:47-68.
[20] Carskadon MA, Acebo C, Jenni OG. Regulations of adolescent sleep: Implications of behaviors. Ann N Y Acad Sci 2004;1021:276-91.
[21] Adam EK, Doane LD, Zinbarg RE, et al. Prospective prediction of major depression disorder from cortisol awakening response in adolescence. Psychoneuroendocrinology 2010;36:921-31.
[22] Boyle SH, Surwit RS, Georgiades A, et al. Depressive symptoms, race, and glucose concentrations. Diabetes Care 2007;30:2484-8.
[23] Matthews K, Schwartz J, Cohen S, et al. Diurnal cortisol decline is related to coronary calcification: CARDIA study. Psychom Med 2006;68:657-61.
[24] Sephton SE, Sapolsky RM, Kraemer HC, Spiegel D. Diurnal cortisol rhythm as a predictor of breast cancer survival. J Natl Cancer Inst 2000;92:994-1000.
[25] Eysenck SB, Eysenck HJ, Barrett P. A revised version of the psychoticism scale. Person Individ Diff 1985;6:21-9.
[26] Lynn R, Martin T. Gender differences in extraversion, neuroticism, and psychoticism in 37 nations. J Soc Psychol 1997;137:369-73.
[27] Sadeh A, Acebo C. The role of actigraphy in sleep medicine. Sleep Med Rev 2002;6:113-24.
[28] Clements AD, Parker CR. The relationship between salivary cortisol concentrations in frozen versus mailed samples. Psychoneuroendocrinology 1998; 23:613-6.
[29] Dressendorfer RA, Kirschbaum C, Rhode W, et al. Synthesis of a cortisolbiotin conjugate and evaluation as a tracer in an immunoassay for salivary cortisol measurement. J Steroid Biochem Mol Biol 1992;43:683-92.
[30] Oakley NR. Validation with polysomnography of the sleepwatch sleep/ wake scoring algorithm used by the actiwatch activity monitoring system. Bend: Mini-Mitter, Cambridge Neurotechnology, 1997.
[31] Littner M, Kushida CA, Anderson WM, et al. Practice parameters for the role of actigraphy in the study of sleep and circadian rhythms: An update for 2002. An American Academy of Sleep Medicine Practice Parameters. Standards of Practice Committee of the American Academy of Sleep Medicine. Sleep 2003;26:337-41.
[32] Sadeh A, Hauri PJ, Kripke DF, Lavie P. The role of actigraphy in the evaluation of sleep disorders. Sleep 1995;18:288-302.
[33] Raudenbush SWB, Bryk AS, eds. Hierarchical linear models: Applications and data analysis methods. Thousand Oaks, CA: Sage Publications, 2002.
[34] Adam EK, Hawkley LC, Kudielka BM, et al. Day-to-day dynamics of experi-ence-cortisol associations in a population-based sample of older adults. Proc Natl Acad Sci USA 2006; 103:17058-63.
[35] Doane LD, Adam EK. Loneliness and cortisol: Momentary, day-to-day, and trait associations. Psychoneuroendocrinology 2010;35:430-41.
[36] Enders CK, Tofighi D. Centering predictor variables in cross-sectional multilevel models: A new look at an old issue. Psychol Methods 2007;12:12138.
[37] Nicolson NA. Measurement of cortisol. In: Luecken LJ, Gallo LC, eds. Handbook of Physiological Research Methods in Health Psychology. Thousand Oaks, CA: Sage Publications, 2008:37-74.
[38] Adam EK, Kumari M. Assessing salivary cortisol in large-scale, epidemiological research. Psychoneuroendocrinology 2009;34:1423-36.
[39] Vazquez-Palacios G, Retana-Marquez S, Bonilla-Jaime H, Velázquez-Moctezuma J. Further definition of the effect of corticosterone on the sleep-wake pattern in the male rat. Pharmacol Biochem Behav 2001;70:305-10.
[40] Holsboer F, von Bardeleben U, Steiger A. Effects of intravenous corticotrop-in- releasing hormone upon sleep-related growth hormone surge and sleep EEG in man. Neuroendocrinology 1998;48:32-8.


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