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Sexual orientation and diurnal cortisol patterns in a cohort of U.S. young adults

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Abstract

Sexual minorities in the United States are at elevated risk of bullying, discrimination, and violence victimization, all stressors that have been linked to psychological and behavioral stress responses including depressive and anxious symptoms and substance use. Acute and chronic stressors may also elicit physiologic stress responses, including changes in the regulation of the hypothalamic-pituitary-adrenocortical (HPA) axis. Few studies, however, have examined the relationship between minority sexual orientation and diurnal cortisol patterns. The present study included 1670 young adults ages 18–32 years (69% female, 31% male) from the Growing Up Today Study, a

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Conflict of interest

The authors have no conflicts of interest to declare.

Contributors

SB Austin, M Rosario were responsible for study conceptualization. SB Austin, M Rosario, KA McLaughlin, AL Roberts, S Missmer, and L Anatale-Tardiff were responsible for data collection. SB Austin, M Rosario, KA McLaughlin, AL Roberts, AR Gordon, V Sarda, S Missmer, L Anatale-Tardiff, and EA Scherer were responsible for data analysis, interpretation, and article preparation.

prospective cohort of U.S. youth. Participants provided five saliva samples over one day to estimate diurnal cortisol patterns. Sexual orientation groups included: completely heterosexual with no same-sex partners (referent), completely heterosexual with same-sex partners/mostly heterosexual, and gay/lesbian/bisexual. Covariates included perceived stress and stressful life events in the past month. Sex-stratified multilevel models of log-transformed cortisol values were used to model diurnal cortisol patterns, and generalized estimating equations were used to model area under the curve (AUC), both with respect to ground (AUCg) and increase (AUCi). Among females, sexual minorities reported significantly more stressful life events in the past month than their heterosexual counterparts. In adjusted multilevel models, sexual orientation was not significantly associated with diurnal cortisol patterns or with AUCg or AUCi in either females or males. There were no significant interactions between sexual orientation and stressful life events. Time-varying negative mood was significantly associated with higher cortisol levels across the day for both female and male participants, after adjusting for all covariates. This study from a large cohort of U.S. young adults did not detect a relationship between sexual orientation and diurnal cortisol patterns. Despite consistent evidence indicating that, compared to heterosexuals, sexual minorities experience elevated exposure to multiple forms of stressors and adversity across the life course, we did not find differences in diurnal cortisol rhythms by sexual orientation. One possible explanation is that sexual minority participants in the study exhibited physiologic resilience.

Keywords

Sexual orientation; Cortisol; Diurnal rhythm; HPA axis; Stressful life events; Young adults

1. Introduction

Health disparities related to sexual orientation are well-documented in the United States, with multiple studies finding sexual minorities (e.g., lesbian, gay, bisexual [LGB], and mostly heterosexual populations) reporting more depressive and anxious symptoms, post-traumatic stress disorder (PTSD), substance use, eating disorder symptoms, and other health risk indicators relative to heterosexual populations (Austin et al., 2013; Institute of Medicine, 2011; Ward et al., 2014). Sexual minority stress theory posits that health inequities may be explained by societal stigmatization of sexual minorities, resulting in differential exposures to acute and chronic stressors such as discrimination, abuse, and violence (Meyer, 2003; Rosario et al., 2002). In support of this theory, elevated rates of violence victimization (Austin et al., 2008; Berlan et al., 2010; Friedman et al., 2011; Katz-Wise and Hyde, 2012; Roberts et al., 2010a; Saewyc et al., 2006) and discrimination (Hatzenbuehler et al., 2015, 2010) are well-documented and have been linked to psychological and behavioral outcomes (Almeida et al., 2009b; Roberts et al., 2012, 2013a). Research on physiological markers of chronic stress will be essential to understanding the mechanisms underlying health disparities—and, more broadly, to understanding the ways that stigmatizing or hostile social environments may “get under the skin” (Hatzenbuehler et al., 2009). Relatively few studies, however, have examined markers of stress physiology across sexual orientation groups.

Chronic stressors can affect health via several pathways, including through psychological responses, coping-related risk behaviors, and neurobiological disruptions. One of the most widely studied physiological stress pathways involves the hypothalamic-pituitary-adrenocortical (HPA) axis, which governs the release of cortisol. Threats to physical and social self-preservation have been shown to influence HPA-axis regulation in numerous studies (Adam and Gunnar, 2001; Dickerson and Kemeny, 2004; Miller et al., 2007). Cortisol has a diurnal rhythm that typically peaks shortly after awakening in the morning (known as the cortisol awakening response [CAR]) and then gradually drops throughout the day. Dysregulation of diurnal cortisol rhythms may result from exposure to chronic stressors (Almeida et al., 2009a; Miller et al., 2007). For instance, experiences of adversity may be associated with hypocortisolism in adulthood, manifesting as low morning cortisol levels and shallow decreases across the day (Gunnar and Vazquez, 2001). Because cortisol is critical to regulation of other physiological systems, including metabolic and immune systems (Dickerson and Kemeny, 2004), cortisol dysregulation may have wide-ranging impacts on pathophysiology (Heim et al., 2000a; Pervanidou and Chrousos, 2012; Segerstrom and Miller, 2004). Specifically, the CAR has also been linked to psychological wellbeing and physical health status, although it not clear whether positive outcomes are associated with larger or smaller CARs (Clow et al., 2004). Flatter cortisol slopes over the course of the day have been associated with PTSD in adults (e.g., (Yehuda et al., 1996)), a history of internalizing disorders in older adolescents (e.g., (Doane et al., 2013)), and chronic conditions such as fibromyalgia and rheumatoid arthritis (Heim et al., 2000a).

Empirical data show clear disparities in victimization and discrimination adversely affecting sexual minorities (Austin et al., 2008; Berlan et al., 2010; Friedman et al., 2011; Hatzenbuehler et al., 2015, 2010; Katz-Wise and Hyde, 2012; Roberts et al., 2010a; Saewyc et al., 2006), and theoretical work offers plausible pathways through which minority sexual orientation might be linked to HPA-axis dysregulation (Meyer, 2003; Rosario et al., 2002). Given that hypocortisolism, characterized by low morning cortisol levels and a blunted decline across the day, is the most common pattern observed among people exposed to adversity (Gunnar and Vazquez, 2001; Heim et al., 2000a), such patterns might be expected among sexual minority populations; however, evidence from sexual minority samples is lacking. To date, only a handful of studies including sexual minorities have assessed cortisol reactivity in experimental settings or diurnal cortisol. One study of gay and bisexual adult men found that degree of “outness” (sexual orientation disclosure) at work was related to higher mean cortisol levels over the course of a work day (Huebner and Davis, 2005). A study of HPA-axis reactivity to a laboratory stressor found LGB young adults who grew up in states classified as highly stigmatizing to sexual minorities as adolescents had a reduced cortisol response to a stress task compared to those who grew up in states classified as having low levels of stigma (Hatzenbuehler and McLaughlin, 2014). This finding reflects prior research showing lower cortisol response following an acute stressor among individuals exposed to adversities in adolescence (Bosch et al., 2012) or in earlier life (Carpenter et al., 2007, 2011). However, much remains to be understood about what distinguishes adaptive from maladaptive cortisol responses and how these responses might be affected by other stressors or buffers relevant for sexual minority populations. For example, another study based on the same LGB sample as used in the Hatzenbuehler and

McLaughlin (2014) study above found that participants with greater parental support—a potential buffer against stigma—had a reduced cortisol response relative to those with less parental support (Burton et al., 2014).

Notably, none of the above studies included a comparison group of heterosexuals. One study of LGB and heterosexual adults in Montreal found no significant differences in diurnal cortisol profiles by sexual orientation identity, although within the LGB group, sexual identity disclosure was associated with lower cortisol levels at 30 min after waking (Juster et al., 2013) A study of HPA axis reactivity in this same sample found differences by sexual orientation and gender, such that lesbian/bisexual women had higher cortisol reactivity following exposure to a laboratory stressor than heterosexual women while gay/bisexual men showed lower overall cortisol levels throughout the study than heterosexual men (Juster et al., 2015). In addition, one study examined associations between sexual orientation identity and other stress-related biomarkers in a nationally representative cohort of U.S. young adults (Everett et al., 2014; Hatzenbuehler et al., 2013). Biomarkers of inflammation and poorer immune function (C-reactive protein and Epstein-Barr virus antibodies) that may be consequences of HPA-axis activation were found to be at higher levels in sexual minority men than in both heterosexual men and sexual minority women (Everett et al., 2014). These studies underscore the importance of considering the intersectionality (Bauer, 2014) of both gender and sexual orientation when examining stress biomarkers.

New research is needed to build a more complete understanding of the relationships among sexual orientation, exposure to stressors, and HPA-axis regulation. In addition, studies are needed that include less well-understood subgroups of sexual minorities such as mostly heterosexuals and heterosexuals with same-sex sexual partners, both of which have been shown to have elevated risk of trauma compared to heterosexuals with no same-sex partners (Roberts et al., 2010a, 2012). The present study sought to address these gaps by examining diurnal patterns of salivary cortisol by gender and sexual orientation in a large epidemiologic cohort of U.S. young adults. Hypotheses of this study were:

1. Compared to heterosexual young adults, sexual minorities will exhibit lower morning levels of diurnal cortisol and shallower decreases across the day;
2. Higher rates of stressful life events among sexual minorities compared to heterosexuals will partly explain differences in diurnal cortisol dysregulation.

2. Methods

2.1. Study sample

Study participants were drawn from the Growing Up Today Study (GUTS), a national, prospective cohort of 27,324 youth. GUTS participants, who were ages 9–16 years at enrollment, are children of women in the Nurses' Health Study 2, a prospective cohort of over 116,000 U.S. women. The first stage of the cohort (GUTS1) began in 1996, followed by a second stage (GUTS2) in 2004. Surveys have been administered annually or biennially since the inception of the cohort. The sample is primarily white (94%) and has a restricted socioeconomic range, as all participants' mothers have a four-year nursing degree.

The current study is based on a subset of GUTS youth who participated in the 2011–2014 GUTS Saliva Substudy, designed to examine sexual orientation in relation to stress response physiology. Only those who had responded to the previous GUTS survey wave in 2010–2011 were invited to participate. Sample selection for the GUTS Saliva Substudy was designed to achieve sufficient subgroup sizes of sexual minorities. All sexual minority participants (i.e., participants who identified as mostly heterosexual, bisexual, mostly homosexual, gay, lesbian on the 2007 or 2010–2011 waves, or reported any history of same-sex sexual partners on the 2005, 2007 or 2010–2011 waves) were recruited. Participants were invited to the GUTS Saliva Substudy by email and then screened for eligibility. Youth were considered ineligible if pregnant currently or in previous six months or if they reported past month use of oral or inhaled steroids, any history of cancer treatment, or ever diagnosed with an endocrine disorder. In total, 6980 GUTS participants were invited, of whom 1966 (28%) took part. Of these, 1686 individuals returned at least one usable saliva sample. We excluded individuals from analysis who were missing information on wakeup time (n = 16), resulting in an analytic sample of 1670 participants. Among the 1966 who participated in the GUTS Saliva Substudy, compared to those included in analyses, those excluded because they did not return at least one usable saliva sample (n = 280) were more likely to be heterosexual, of white ethnicity, and older ($P < 0.05$) but did not differ by sex ($P > 0.05$) compared to those included.

2.2. Survey measures

2.2.1. Sexual orientation—Sexual orientation was assessed on the GUTS 2005, 2007, 2010–2011, and 2013 survey waves using two widely used measures. The first measure asked participants to report which of the following best describes them: Completely heterosexual (attracted to persons of the opposite sex); mostly heterosexual; bisexual (equally attracted to men and women); mostly homosexual; completely homosexual, gay, or lesbian (attracted to persons of the same sex); or unsure (Remafedi et al., 1992). The second asked participants to report the sex of any sexual partners they have had (female[s], male[s], female[s] and male[s], or no sexual contact). Due to sample size limitations, sexual orientations groups were aggregated for analysis into three groups: (1) completely heterosexual with no same-sex partners, (2) mostly heterosexual or completely heterosexual with same-sex partners, and (3) lesbian, gay, or bisexual. Prior research (Roberts et al., 2010b), including in GUTS (Roberts et al., 2012), indicates that completely heterosexuals with same-sex partners and mostly heterosexuals have elevated health risks compared to completely heterosexuals with no same-sex partners, so the former two groups were combined to make an additional category for analyses. Sexual orientation was carried forward from the most recent prior wave for participants missing sexual orientation on the 2013 wave.

2.2.2. Recent stressful life events—The GUTS Saliva Substudy included a daily diary and brief questionnaire that participants completed on the day of saliva collection. This questionnaire included a modified version of the Stressful Life Events Screening Questionnaire (Goodman et al., 1998) assessing self-reported lifetime exposure to 13 types of traumatic experiences (e.g., life-threatening events, death of a loved one). To capture a full breadth of stressful life experiences, the original measure was modified by adding four

questions about stressful life events pertaining to financial hardship and divorce or separation. In addition, to ascertain recent exposures to stressors, the time frame was modified to focus on experiences in the past 30 days and past year. One binary variable was created indicating whether any of the stressful events had been experienced within the past 30 days (any vs. none) and a second variable indicated any experiences in the past year.

2.2.3. Daily diary measures and additional covariates—The saliva diary included measures accounting for factors that may impact diurnal rhythms (Adam and Kumari, 2009). These included measures of sleep duration and quality (time at falling asleep on night before data collection, time of awakening on day of data collection, previous night's hours of sleep, and usual hours of sleep) and mood during each saliva collection ("Do you feel worried, anxious, or fearful right now?" and "Do you feel happy, excited, or content right now?" with response options from (1) Not at all to (4) Extremely). The "worried" and "happy" mood items were included as ordinal time-varying covariates in the model, with higher scores indicating greater levels of worry or happiness at each time point. The saliva diary also included the four-item Perceived Stress Scale (PSS-4) (Cohen et al., 1983), which asked participants to report feelings and thoughts in the previous month (e.g., "In the last month, how often have you felt you were unable to control the important things in your life?" with response options from (0) Never to (4) Very often) (Cohen et al., 1983; Cohen and Williamson, 1988). PSS-4 scores were obtained by reverse coding the two positive items and taking the mean of the four items, such that higher scores indicated higher perceived stress.

Participants were asked to report whether they had, on the day of saliva collection, smoked any cigarettes (yes/no), had any alcoholic beverages (yes/no), engaged in any vigorous physical activity (increasing heart rate and sweat; yes/no), and to list any drugs or medications taken. Women were asked if they were currently using or had used in the previous month any form of hormonal contraception (yes/no). Additional covariates included participant age (in years) and non-compliance with the wakeup sample (yes/no; non-compliance defined as the first saliva sample not being taken within 15 min of waking up).

2.3. Salivary cortisol collection

Participants received saliva collection tubes through the mail along with written instructions and the daily diary. Samples were collected from participants on a single weekday (Monday–Thursday). After completing the informed consent process, participants were instructed to provide five passive-drool saliva samples over the course of the day: at awakening, 45 min, 4, and 10 h after awakening, and at bedtime. Participants were instructed not to brush their teeth before the first (awakening) sample was collected and not to eat, drink, chew anything or do vigorous physical activity for at least 30 min before each subsequent sample. To assess adherence, participants recorded the time of collection for each sample and whether or not they had brushed teeth, eaten, drunk, or exercised in the previous 30 min. Filled tubes were stored in baggies in a refrigerator in the participant's home until sampling was complete and then returned along with an ice pack via two-day delivery in a postage-paid mailer. All saliva samples were collected between August 2011

and February 2014. Participants received $25 upon return of saliva samples. This study was approved by the Brigham & Women's Hospital Institutional Review Board.

2.4. Cortisol processing

Saliva samples were shipped to the Channing Division of Network Medicine (CDNM) biorepository, aliquoted, stored at −20 °Centigrade, and assayed by Rohleder Lab at Brandeis University along with quality control (QC) samples provided by the CDNM. The CDNM biorepository routinely stocks pooled QC samples. Three distinct QC pools of saliva were obtained. Aliquots of QC pools were distributed randomly among the participant samples and labeled with alias IDs for blinding. Each batch of samples analyzed contained two QC pool samples from at least two of the three available pools. Each pool was represented in at least one-third of the batches analyzed. At Rohleder Lab, cortisol was measured in duplicate using a competitive chemiluminescence immunoassay (CLIA; IBL-International, Toronto, ON, Canada). Inter- and intra-assay coefficients of all assays were below 10%. Rohleder Lab provided CDNM with final data from study samples and QC samples, which were un-blinded and QC results were compiled and analyzed across the study. Coefficients of variation for the study met the biorepository standard of 15% or below (observed range: 9.7%–13.3%). Batch adjustment was performed to adjust for any potential batch effects (Rosner et al., 2008).

2.5. Statistical analysis

Multilevel models of log-transformed cortisol values were used to examine diurnal cortisol patterns (interpreted in nmol/L). The models included individual-level, day-level, and sample-level covariates in addition to individual-level random intercept (awakening cortisol level) and slope terms (time since waking). All continuous covariates were centered around their mean. The primary predictor was sexual orientation, with completely heterosexual without same-sex partners as the referent group. In addition to sexual orientation, the models included a linear, quadratic, and cubic effect of time since waking and an indicator variable for non-compliance with the wake-up sample time (defined as >15 min from wake-up time). Models also included an estimate of cortisol awakening response (CAR) and an interaction between CAR and sexual orientation to test whether the CAR differed by sexual orientation. CAR is defined as the difference between the wake-up sample (Sample 1) and the sample collected 45 min later (Sample 2). Only those covariates that were either associated with cortisol or sexual orientation were included in final models. As models were of log-transformed values of cortisol, parameter estimates can be transformed (as $100*(\exp(\beta) - 1)$ for interpretation as percentage increase or decrease in mean cortisol value (Adam et al., 2006). All analyses were sex-stratified so that models for females could account for hormonal contraception use.

In models examining the effect of stressful events on cortisol, a main effect of stressful events and an interaction of stressful events with sexual orientation were additionally included in the model to determine whether stressful events influenced cortisol level and whether this effect differed by sexual orientation. We also calculated cortisol area under the curve (AUC), both with respect to ground (AUCg) to assess overall cortisol output and with respect to increase (AUCi) to assess changes in cortisol output over time (Fekedulegn et al.,

2007; Pruessner et al., 2003). Generalized estimating equations were used to estimate associations between sexual orientation and AUCg/AUCi. Models were adjusted for covariates that were either associated with AUCg/AUCi or sexual orientation. All analyses were conducted using SAS v9.3 (SAS, 2012)

3. Results

3.1. Descriptive statistics

The analytic sample included 1152 (68.9%) female participants and 518 (31.1%) male participants ages 18–32 years old. Approximately 8% of the sample identified as bisexual, gay, or lesbian (n = 130), 26% of the sample identified as mostly heterosexual or completely heterosexual with same-sex partners (n = 427; hereafter referred to as “mostly heterosexual”) and 67% of the sample identified as completely heterosexual and had no same-sex partners (n = 1113). At the bivariate level, sexual minority status was significantly associated with reported stressful life events in the past month for female participants. Male sexual minorities reported somewhat more stressful life events than heterosexuals, but this difference did not reach statistical significance (Table 1). Among females, 15% of completely heterosexual participants reported two or more stressful events in the past year, compared to 19% of mostly heterosexual and 31% of lesbian or bisexual participants (p = 0.001). Among males, 16% of completely heterosexual, 25% of mostly heterosexual, and 20% of bisexual or gay participants reported two or more stressful events in the past month (p = 0.13).

3.2. Diurnal cortisol by sexual orientation

As depicted in Fig. 1, on average, salivary cortisol concentration followed the expected diurnal pattern, after accounting for key covariates. Participants exhibited high mean cortisol values upon awakening (Female (F): 15.8 nmol/L; Male (M): 16.0 nmol/L), an increase in concentration in the first 45 min after awakening (F: 48% increase; M: 39% increase), and decrease across the rest of the day (F: 15% lower and M: 14% lower per hour than at wakeup) (Table 2 for females and Table 3 for males).

Sexual orientation was not significantly associated with diurnal cortisol for females or males (see Tables 2 and 3). There was no significant interaction between sexual orientation and stressful life events in the past 30 days on diurnal cortisol for females or males (Table 4 for females and Table 5 for males), and results were similar for past-year stressful life events (data not shown). Time-varying negative mood (self-reported worry/anxiety at each saliva collection) was significantly associated with increased mean cortisol levels across the day for both female and male participants, after adjustment for recent stressful life events.

Table 6 presents results of generalized estimating equations examining sexual orientation as a predictor of overall cortisol output as measured by AUCg, adjusted for covariates. Sexual orientation was not significantly associated with AUCg; findings for AUCi were similar (data not shown).

4. Discussion

To our knowledge, this is the largest study to date to examine associations between sexual orientation and any marker of HPA-axis functioning, and one of the larger studies of diurnal cortisol patterns in young adults in general. We did not find support for a hypothesized association between sexual orientation and diurnal cortisol rhythms. We observed a statistically significantly higher prevalence of stressful life events in sexual minority females compared to heterosexual females, as would be predicted by sexual minority stress theory, though only a suggestion of higher prevalence among sexual minority males compared to heterosexual males. We did not find evidence of an interaction between stressful life events and sexual orientation.

These findings represent an important contribution to the literature on social stressors in relation to stress physiology. Most prior research on sexual orientation and stress biomarkers has focused on within-group analyses of sexual minorities without a heterosexual comparison group (Burton et al., 2014; Hatzenbuehler et al., 2014; Huebner and Davis, 2005). The only other study to assess diurnal cortisol and sexual orientation with a heterosexual comparison group also found no significant associations between sexual orientation identity and cortisol profile in a sample of 87 LGB and heterosexual adults (Juster et al., 2013).

What might explain the lack of differences by sexual orientation in cortisol diurnal rhythms in young adults? One possible explanation could be that the similarity across sexual orientation groups may be the result of physiologic resilience in sexual minority participants in the study. Sexual minority youth and adults are exposed to both chronic and acute social stressors related to stigma and discrimination (Institute of Medicine, 2011). Indeed, sexual minority female participants in this study reported a higher prevalence of recent stressful life events relative to completely heterosexuals. Researchers have proposed that sexual minorities, particularly those who have access to supportive social contexts, may be able to develop adaptive coping strategies that heighten resilience (Hatzenbuehler et al., 2009). Following from this logic, it is possible that young sexual minority adults in the GUTS cohort have developed coping strategies that enhance their ability to manage stigma-related stressors and are reflected in HPA-axis responses that parallel those of their heterosexual peers. Prior research on stress reactivity finding low cortisol response to a laboratory stressor among gay/bisexual men in relation to heterosexual men was also hypothesized to be due to successful adaptation to sexual minority stigma (Juster et al., 2015). That said, the well-documented higher rates of depressive symptoms, PTSD, eating disorder symptoms, substance use, and other health risk indicators found in sexual minority compared to heterosexuals in the GUTS cohort (Austin et al., 2013; Corliss et al., 2010; Roberts et al., 2012, 2013b) raises the question as to whether coping strategies that mitigate dysregulation of the HPA axis or, more particularly, cortisol diurnal rhythms, may be different from strategies that mitigate mental health problems.

Another possible explanation may be that diurnal cortisol rhythms may be driven more by proximal emotions than by past stressful events. Most of the evidence for disruptions in cortisol regulation among adults with abuse histories or among LGB adults who grew up in

high-stigma or low-family-support environments is based on reactivity to an acute stressor or challenge administered in a laboratory setting (Burton et al., 2014; Hatzenbuehler and McLaughlin, 2014; Heim et al., 2008, 2000b, 2002). In the present study, self-reported current level of worry/anxiety, which did not exhibit large variations by sexual orientation, was one of the few covariates that was significantly associated with diurnal cortisol levels, which is consistent with findings that feelings such as anger/tension predict higher evening cortisol levels (Adam et al., 2006) and that daily stressors, such as arguments at home, predict increased total cortisol output (Stawski et al., 2013)

The nature and timing of the stressor also play a critical role in diurnal cortisol patterns, according to a meta-analysis of studies on stress and HPA-axis functioning (Fekedulegn et al., 2007). For example, traumatic stress or stress due to physical threat, like combat, produces a high and flat diurnal cortisol pattern, while stress that threatens the social self, such as divorce, produces significantly higher cortisol secretion at certain times of day. In the present study, although we did not see associations between past-month stressful life events and cortisol patterns, if different types of life events produce conflicting diurnal cortisol profiles, then the analyses presented here would not be able to detect these relationships. Future research should investigate the role of specific types of stressors (e.g., physical threats such as overt violence versus social-evaluative threats such as covert employment discrimination) in the diurnal cortisol patterns of LGB and heterosexual populations. Finally, previous research has found associations between diurnal cortisol patterns and sexual identity milestones (e.g., sexual orientation disclosure) (Huebner and Davis, 2005; Juster et al., 2016, 2013). Although outside the scope of the present study, further research is needed to examine how such processes may or may not be protective against HPA-axis dysregulation.

These findings must be considered in light of several limitations. In spite of the large sample size, power limitations are a concern for this analysis, particularly given the relatively small sample of LGB individuals who provided saliva for the study, made even smaller when stratified by sex. Future studies with even larger samples are needed because, as indicated by prior research on stress-related biomarkers (Everett et al., 2014; Juster et al., 2015, 2013) and guided by intersectionality theory (Bauer, 2014), it is critically important to be able to examine the intersection of sexual orientation and gender to improve understanding of physiological stress response. Larger samples and oversampling of transgender participants would also allow for analyses of gendered patterns beyond those seen in cisgender populations. We did not monitor compliance with sampling protocols electronically, although we did capture self-reported sampling times and compliance with instructions (e.g., not eating or drinking 30 min prior), and models were adjusted for sampling-time noncompliance. Furthermore, the low response rate may have introduced bias in our findings. Finally, participants were members of an ongoing cohort of the children of U.S. nurses and are predominantly white and raised in largely middle-income households; thus, these findings may not be generalizable to other young adult populations.

5. Conclusion

Given the well-documented health inequities faced by U.S. sexual minority populations and mounting evidence that social stressors such as experiences of stigma and discrimination can influence both mental and physical health, this study offers a needed contribution to the research literature on the dynamics of stress biomarkers in relation to the health of socially marginalized populations. If future studies replicate our results, that will suggest that diurnal cortisol dysregulation, at least as currently conceived and measured, may not be on a pathway linking sexual minority stressors to the widely observed elevated risk of depressive and anxious symptoms, PTSD, eating disorders, and substance use among sexual minority adolescents and young adults. Future research on sexual orientation and the HPA axis must also examine specific types of social stressors, given the dependence of cortisol patterns on the specific type and timing of the stressor. Finally, given the rapidly shifting status of civil rights for sexual minority individuals in the United States, longitudinal research on the relationship between sexual orientation and stress biomarkers may aid in evaluating whether and how changes in the social environment may impact individual physiological stress processes.

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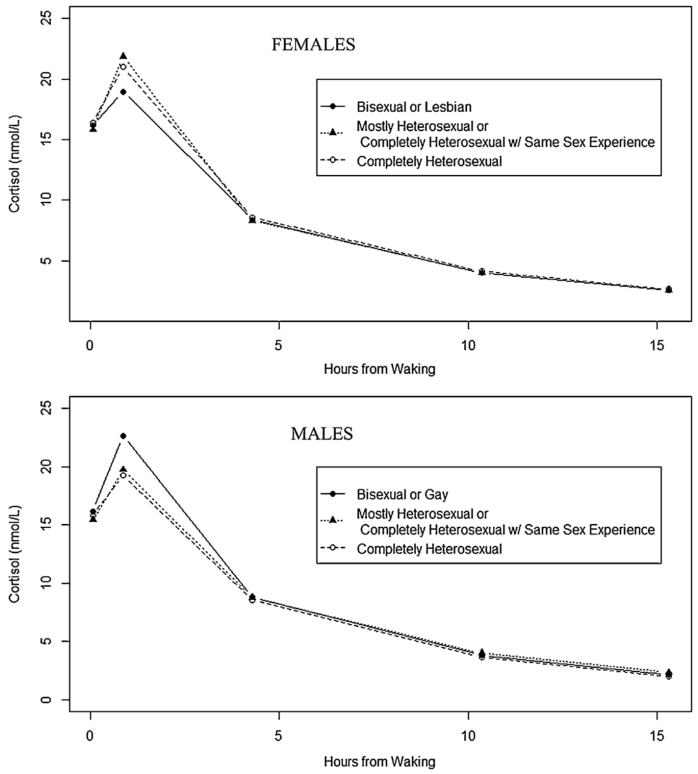

Fig. 1.

Estimated model-based mean cortisol values as a function of time since awakening, by sexual orientation among female and male participants ages 18–32 years in the Growing Up Today Study (GUTS) Saliva Substudy. Notes: estimates based on model presented in Table 3. Saliva sample times: sample 1 = 0 from waking; sample 2 = 45 min from waking; sample 3 = 4 from waking; sample 4 = 10 from waking; sample 5 = before bed. No significant differences in cortisol were seen across sexual orientation groups.

Table 1

Participant characteristics for females and males in the Growing Up Today Study who completed saliva sampling in 2011–2014 by sexual orientation identity (n = 1670).

	FEMALES				MALES			
	Completely heterosexual (CH) (n = 758)	**Mostly heterosexual or CH w/SS partners (n = 324)**	**Bisexual or lesbian (n = 70)**	***p***	**Completely heterosexual (CH) (n = 362)**	**Mostly heterosexual or CH w/SS partners (n = 95)**	**Bisexual or gay (n = 61)**	***p***
Age in years, *M* (SD)	24.4 (3.5)	25.1 (3.1)	24.9 (3.6)	0. 01	24.8 (3.5)	25.1 (3.3)	24.7 (3.1)	0.63
Sleep, *M*(SD)								
Hours of sleep in previous night	7.8 (1.3)	7.6 (1.3)	7.7 (1.5)	0.21	7.3 (1.4)	7.4 (1.4)	7.5 (1.3)	0.70
Hours of sleep on typical night	7.7 (0.9)	7.7 (1.0)	7.8 (0.9)	0.59	7.4 (1.0)	7.5 (0.9)	7.4 (0.8)	0.67
Waking time (HH:MM)	7:57 AM (1:36)	7:59 AM (1:37)	8:13 AM (1:38)	0.43	7:57 AM (1:46)	8:03 AM (1:44)	8:16 AM (2:12)	0.43
Health-related behaviors, n (%)								
Any alcohol use	164 (21.9)	104 (32.6)	15 (21.4)	<0.001	94 (26.6)	32 (34.4)	22 (37.3)	0.12
Any cigarette smoking	14 (1.9)	17 (5.3)	5 (7.1)	0.002	17 (4.7)	7 (7.4)	5 (8.2)	0.39
Hormonal contraception use (female only), *n* (%)	389 (51.6)	172 (53.3)	22 (31.4)	0.003	n/a	n/a	n/a	
Perceived stress scale, *M* (SD)	2.2 (0.7)	2.4 (0.7)	2.5 (0.7)	>.001	2.2 (0.7)	2.5 (0.8)	2.5 (0.8)	<0.001
Time-varying worry score, *M* (SD)								
Sample 1 (Wake up)	1.5 (0.6)	1.5 (0.6)	1.5 (0.6)	0.13	1.4 (0.5)	1.5 (0.6)	1.5 (0.6)	0.02
Sample 2 (45 min post-wake up)	1.5 (0.6)	1.5 (0.6)	1.5 (0.6)	0.69	1.4 (0.5)	1.5 (0.6)	1.5 (0.6)	0.06
Sample 3 (4 post-wake up)	1.5 (0.6)	1.6 (0.6)	1.5 (0.6)	0.05	1.4 (0.6)	1.5 (0.6)	1.5 (0.6)	0.03
Sample 4 (10 post-wake up)	1.5 (0.6)	1.6 (0.7)	1.6 (0.7)	0.04	1.4 (0.6)	1.6 (0.7)	1.6 (0.7)	0.03
Sample 5 (Before bed)	1.5 (0.6)	1.6 (0.7)	1.7 (0.7)	0.002	1.4 (0.6)	1.6 (0.7)	1.4 (0.7)	0.01
Time-varying happy score, *M* (SD)								
Sample 1 (Wake up)	2.0 (0.7)	1.8 (0.6)	1.9 (0.5)	0.001	1.9 (0.7)	1.9 (0.7)	1.8 (0.6)	0.66
Sample 2 (45 min post-wake up)	2.1 (0.6)	2.0 (0.6)	2.1 (0.6)	0.05	2.1 (0.6)	2.0 (0.7)	2.0 (0.5)	0.38
Sample 3 (4 post-wake up)	2.3 (0.7)	2.1 (0.6)	2.1 (0.7)	<0.001	2.3 (0.7)	2.1 (0.7)	2.2 (0.6)	0.02
Sample 4 (10 post-wake up)	2.4 (0.7)	2.2 (0.7)	2.2 (0.6)	0.02	2.3 (0.7)	2.1 (0.8)	2.2 (0.7)	0.11
Sample 5 (Before bed)	2.2 (0.7)	2.1 (0.7)	2.1 (0.6)	0.18	2.3 (0.8)	2.0 (0.7)	2.1 (0.7)	0.03
Stressful events in past month, *n* (%)				0.001				0.25
None	643 (84.8)	261 (80.6)	48 (68.6)		301 (83.2)	72 (75.8)	49 (80.3)	
1+	115 (15.2)	63 (19.4)	22 (31.4)		61 (16.9)	23 (24.2)	12 (19.7)	

	FEMALES				MALES			
	Completely heterosexual (CH) (n = 758)	**Mostly heterosexual or CH w/SS partners (n = 324)**	**Bisexual or lesbian (n = 70)**	***p***	**Completely heterosexual (CH) (n = 362)**	**Mostly heterosexual or CH w/SS partners (n = 95)**	**Bisexual or gay (n = 61)**	***p***
Compliant with wakeup sample, *n* (%)	728 (96.7)	306 (94.4)	66 (94.3)	0.18	352 (97.8)	89 (98.9)	60 (98.4)	0.78

Notes: P-values based on Chi-square tests for categorical variables and ANOVA for continuous variables. Compliant with wake up sample = saliva sample taken within 15 min of waking. All numbers are rounded to nearest zero.

Table 2

Multilevel model of log(cortisol) including random effects for wakeup cortisol and time since waking, and fixed effects of cortisol awakening response and time since waking squared and cubed among females in GUTS Saliva Substudy (n = 1113).

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Model for wakeup cortisol level				
Intercept	2.763	0.021	<0.0001	**15.9 nmol/L mean wakeup cortisol** [a]
Age in years	0.030	0.003	<0.0001	**3.0% higher for each year older**
Hours sleep, previous night	−0.031	0.008	0.0001	**3.1% lower for each hour less sleep**
Hours sleep, typical night	−0.015	0.010	0.16	−1.5% (n.s.)
Waking time	0.000	0.000	<0.0001	**0.001% lower per minute later of wakeup time**
Any cigarette smoking	0.100	0.053	0.06	10.5% (n.s.)
Any alcoholic drinks	0.006	0.021	0.80	0.6% (n.s.)
Perceived stress scale	0.020	0.014	0.14	2.0% (n.s.)
Hormonal contraception use	0.064	0.018	0.0005	**6.6% higher if on hormonal contraception**
Noncompliance (wakeup sample)	0.091	0.053	0.09	9.6% (n.s.)
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.033	0.030	0.27	−3.3% (n.s.)
Bisexual or Lesbian	−0.012	0.056	0.82	−1.2% (n.s.)
Model for cortisol level				
Worry level (at each sample)	0.025	0.013	0.06	2.5% (n.s.)
Happiness level (at each sample)	−0.011	0.012	0.37	−1.1% (n.s.)
Model for time since waking effect				
Intercept	−0.168	0.005	<0.0001	**15.5% lower per hour since wakeup**
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.000	0.004	0.99	−0.0% (n.s.)
Bisexual or Lesbian	−0.002	0.007	0.77	−0.2% (n.s.)
Model for time since waking squared effect				
Intercept	0.003	0.000	<0.0001	**0.4% per hour squared since wakeup**
Model for time since waking cubed effect				
Intercept	−0.000	0.000	<0.0001	**0.0% per hour cubed since wakeup**
Model for cortisol awakening response effect				
Intercept	0.389	0.023	<0.0001	**47.6% higher if CAR**
Noncompliance (wakeup sample)	−0.190	0.088	0.03	**17.3% lower if non-compliant with wakeup sample**
Sexual orientation (Ref = CH)				
MH or CHwSS	0.074	0.042	0.08	7.6% (n.s.)
Bisexual or Lesbian	−0.087	0.078	0.26	−8.3% (n.s.)

Random Effects	Variance estimate	SE	p-value
Awakening cortisol	0.029	0.006	<0.0001

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Time since waking	0.001		0.000	<0.0001

Notes: Variables centered around their means are age, hours sleep in previous night, hours sleep on a typical night, waking time, perceived stress score, level of worry during each saliva sample. Sexual orientation categories: (1) Bisexual or Lesbian (2) MH or CHwSS = Mostly Heterosexual or Completely Heterosexual with same-sex partners; (3) CH = Completely Heterosexual with no same-sex partners (Ref). All numbers are rounded to nearest zero.

[a] Mean wakeup cortisol among CH, non-smoking, no alcohol, no hormonal contraception use, compliant participants with mean age sleep, waking time, stress, and worryscores.

[b] In interpreting effect sizes, use of a logarithmic outcome allows coefficients to be interpreted as percentage change in the outcome per unit change in the independent variable, after applying the following transformation: $B_{\%change} = [\exp(B_{raw})] - 1$. The "Interpretation" column of table includes the percentage change interpretation based on this transformation.

Table 3

Multilevel model of log(cortisol) including random effects for wakeup cortisol and time since waking, and fixed effects of cortisol awakening response and time since waking squared and cubed among males in GUTS Saliva Substudy (n = 487).

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Model for wakeup cortisol level				
Intercept	2.773	0.032	<0.0001	**16.0 nmol/L mean wakeup cortisol** [a]
Age in years	0.030	0.005	<0.0001	**3.0% higher for each year older**
Hours sleep, previous night	0.000	0.013	0.99	0.0% (n.s.)
Hours sleep, typical night	−0.025	0.017	0.15	−2.5% (n.s.)
Waking time	0.000	0.000	0.12	0.0% (n.s.)
Any cigarette smoking	−0.052	0.067	0.44	−5.1% (n.s.)
Any alcoholic drinks	−0.038	0.033	0.25	−3.8% (n.s.)
Perceived stress scale	−0.016	0.023	0.48	−1.6% (n.s.)
Noncompliance (wakeup sample)	0.300	0.124	0.02	**35.0% higher if non-compliant with wakeup sample**
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.023	0.058	0.69	−2.2% (n.s.)
Bisexual or Gay	0.023	0.069	0.74	2.3% (n.s.)
Model for cortisol level				
Worry level (at each sample)	0.055	0.024	0.02	**5.7% higher with increased worry**
Happiness level (at each sample)	−0.022	0.020	0.27	−2.2% (n.s.)
Model for time since waking effect				
Intercept	−0.145	0.010	<0.0001	**13.5% lower per hr since wakeup**
Sexual orientation (Ref = CH)				
MH or CHwSS	0.011	0.007	0.12	1.1% (n.s.)
Bisexual or Gay	0.002	0.008	0.85	0.2% (n.s.)
Model for time since waking squared effect				
Intercept	0.000	0.001	0.78	0.0% (n.s.)
Model for time since waking cubed effect				
Intercept	0.000	0.000	0.03	**0.0% per hour cubed since wakeup**
Model for cortisol awakening response effect				
Intercept	0.321	0.039	<0.0001	**37.8% higher if CAR**
Noncompliance (wakeup sample)	−0.219	0.207	0.29	−19.7% (n.s.)
Sexual orientation (Ref = CH)				
MH or CHwSS	0.038	0.084	0.65	3.9% (n.s.)
Bisexual or Gay	0.139	0.101	0.17	14.9% (n.s.)

Random Effects	Variance estimate	SE	p-value
Awakening cortisol	0.020	0.011	0.04
Time since waking	0.001	0.000	<0.0001

Notes: Variables centered around their means are age, hours sleep in previous night, hours sleep on a typicalnight, waking time, perceived stress score, level of worry during each saliva sample. Sexual orientation categories: (1) Bisexual or Gay (2) MH or CHwSS = Mostly Heterosexual or Completely Heterosexual with same-sex partners; (3) CH = Completely Heterosexual with no same-sex partners (Ref). All numbers are rounded to nearest zero.

[a] Mean wakeup cortisol among CH, non-smoking, no alcohol, compliant participants with mean age sleep, waking time, stress, and worry scores.

[b] In interpreting effect sizes, use of a logarithmic outcome allows coefficients to be interpreted as percentage change in the outcome per unit change in the independent variable, after applying the following transformation: $B_{\%change} = [\exp(B_{raw})] - 1$. The "Interpretation" column of table includes the percentage change interpretation based on this transformation.

Table 4

Multilevel model of log(cortisol) including any stressful life events in the previous month and random effects for wakeup cortisol and time since waking, and fixed effects of cortisol awakening response and time since waking squared and cubed among females in GUTS Saliva Substudy (n = 1113).

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Model for wakeup cortisol level				
Intercept	2.761	0.022	<0.0001	**15.8 nmol/L mean wakeup cortisol** [a]
Age in years	0.030	0.003	<0.0001	**3.0% higher for each year older**
Hours sleep, previous night	−0.030	0.008	0.0002	**3.0% lower for each hour less sleep**
Hours sleep, typical night	−0.016	0.010	0.13	−1.6% (n.s.)
Waking time	0.000	0.000	<0.0001	**0.001% lower per minute later of wakeup time**
Any cigarette smoking	0.098	0.053	0.06	10.3% (n.s.)
Any alcoholic drinks	0.006	0.021	0.77	0.6% (n.s.)
Stress scale	0.014	0.014	0.32	1.4% (n.s.)
Hormonal contraception use	0.0654	0.018	0.0004	**6.7% higher if on hormonal contraception**
Noncompliance (wakeup sample)	0.092	0.053	0.08	9.7% (n.s.)
1+ traumatic events, past month	0.009	0.032	0.78	0.9% (n.s.)
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.045	0.031	0.15	−4.4% (n.s.)
Bisexual or Lesbian	−0.022	0.061	0.72	−2.2% (n.s.)
Sexual orientation by stressful events interaction (Ref = CH w/no past month stressful events)				
1+ stressful events × MH/CHwSS	0.067	0.054	0.21	6.9% (n.s.)
1+ stressful events × Bi/Lesbian	0.032	0.085	0.70	3.3% (n.s.)
Model for cortisol level				
Worry level (at each sample)	0.025	0.013	0.06	**2.5% higher with increased worry**
Happiness level (at each sample)	−0.011	0.012	0.35	−1.1% (n.s.)
Model for time since waking effect				
Intercept	−0.168	0.005	<0.0001	**15.5% lower per hour since wakeup**
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.000	0.004	0.99	−0.0% (n.s.)
Bisexual or Lesbian	−0.002	0.007	0.76	−0.2% (n.s.)
Model for time since waking squared effect				
Intercept	0.003	0.000	<0.0001	**0.4% per hour squared since wakeup**
Model for time since waking cubed effect				
Intercept	−0.00002	0.000004	<0.0001	**0.0% per hour cubed since wakeup**
Model for cortisol awakening response (CAR) effect				

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Intercept	0.389	0.023	<0.0001	**47.6% higher if CAR**
Noncompliance (wakeup sample)	−0.190	0.088	0.03	**17.3% lower if non-compliant with wakeup sample**
Sexual orientation (Ref = CH)				
MH or CHwSS	0.074	0.042	0.08	7.7% (n.s.)
Bisexual or Lesbian	−0.087	0.078	0.26	−8.3% (n.s.)

Random Effects	Variance estimate	SE	p-value
Awakening cortisol	0.029	0.006	<0.0001
Time since waking	0.001	0.000	<0.0001

Notes: Variables centered around their means are age, hours sleep in previous night, hours sleep on a typical night, waking time, perceived stress, worry, happiness. Sexual orientation categories: (1) Bisexual or Lesbian; (2) MH, CHwSS = Mostly Heterosexual or Completely Heterosexual with same-sex partners; (3) CH = Completely Heterosexual with no same-sex partners (Ref). All numbers are rounded to nearest zero.

[a] Mean wakeup cortisol among CH, non-smoking, no alcohol, compliant participants with no reported stressful events in the previous month and mean sleep, waking time, stress, worry, and happiness scores.

[b] In interpreting effect sizes, use of a logarithmic outcome allows coefficients to be interpreted as percentage change in the outcome per unit change in the independent variable, after applying the following transformation: $B_{\%change} = [\exp(B_{raw})] - 1$. The "Interpretation" column of table includes the percentage change interpretation based on this transformation.

Table 5

Multilevel model of log(cortisol) including any stressful life events in the previous month and random effects for wakeup cortisol and time since waking, and fixed effects of cortisol awakening response and time since waking squared and cubed among males in GUTS Saliva Substudy (n = 487).

Variable	Estimate	SE	p-value	Interpretation (% increase/decrease per unit change of predictor)[b]
Model for wakeup cortisol level				
Intercept	2.783	0.033	<0.0001	**16.2 nmol/L mean wakeup cortisol** [a]
Age in years	0.030	0.005	<0.0001	**3.0% higher for each year older**
Hours sleep, previous night	−0.001	0.013	0.95	−0.1% (n.s.)
Hours sleep, typical night	−0.023	0.017	0.19	−2.3% (n.s.)
Waking time	0.000	0.000	0.14	0.0% (n.s.)
Any cigarette smoking	−0.063	0.067	0.35	−6.1% (n.s.)
Any alcoholic drinks	−0.039	0.033	0.25	−3.8% (n.s.)
Perceived stress	−0.010	0.024	0.70	−1.0% (n.s.)
Noncompliance (wakeup sample)	0.305	0.124	0.01	**35.6% higher if noncompliant with wakeup sample**
1+ stressful events, past month	−0.050	0.050	0.31	−4.9% (n.s.)
Sexual orientation (Ref = CH)				
MH or CHwSS	−0.003	0.061	0.96	−0.3% (n.s.)
Bisexual or Gay	−0.009	0.073	0.90	0.0% (n.s.)
Sexual orientation by stressful events interaction (Ref = CH w/no past month stressful events)				
1+ stressful events × MH/CHwSS	−0.078	0.096	0.42	−7.5% (n.s.)
1+ stressful events × Bi/Gay	0.156	0.118	0.19	17.0% (n.s.)
Model for cortisol level				
Worry level (at each sample)	0.056	0.024	0.02	**5.7% higher with increased worry**
Happiness level (at each sample)	−0.022	0.020	0.28	−2.2% (n.s.)
Model for time since waking effect				
Intercept	−0.145	0.010	<0.0001	**13.5% lower per hour since wakeup**
Sexual orientation (Ref = CH)				
MH or CHwSS	0.011	0.007	0.12	1.1% (n.s.)
Bisexual or Gay	0.002	0.008	0.85	0.2% (n.s.)
Model for time since waking squared effect				
Intercept	0.000	0.001	0.79	0.0% (n.s.)
Model for time since waking cubed effect				
Intercept	0.000	0.000	0.03	**0.0% per hour cubed since wakeup**
Model for cortisol awakening response effect				
Intercept	0.321	0.039	<0.0001	**37.8% higher if CAR**
Noncompliance (wakeup sample)	−0.219	0.207	0.29	−19.7% (n.s.)
Sexual orientation (Ref = CH)				

Variable	**Estimate**	**SE**	**p-value**	**Interpretation (% increase/decrease per unit change of predictor)**[b]
MH or CHwSS	0.038	0.084	0.65	3.9% (n.s.)
Bisexual or Gay	0.138	0.101	0.17	14.8% (n.s.)

Random Effects	Variance estimate	SE	p-value
Awakening cortisol	0.020	0.011	0.04
Time since waking	0.001	0.000	<0.0001

Notes: Variables centered around their means are age, hours sleep in previous night, hours sleep on a typical night, waking time, perceived stress, worry, happiness. Sexual orientation categories: (1) Bisexual or Gay; (2) MH, CHwSS = Mostly Heterosexual or Completely Heterosexual with same-sex partners; (3) CH = Completely Heterosexual with no same-sex partners (Ref). All numbers are rounded to nearest zero.

[a] Mean wakeup cortisol among CH, nonsmoking, no alcohol, compliant participants with no reported stressful events in the previous month and mean sleep, waking time, stress, worry, and happiness scores.

[b] In interpreting effect sizes, use of a logarithmic outcome allows coefficients to be interpreted as percentage change in the outcome per unit change in the independent variable, after applying the following transformation: $B_{\%change} = [\exp(B_{raw})] - 1$. The "Interpretation" column of table includes the percentage change interpretation based on this transformation.

Table 6

Adjusted generalized estimating equations (GEE) models of cortisol output (AUCg) by sexual orientation among females and males in GUTS Saliva Substudy (n = 1502).

	FEMALES (n = 1047)			MALES (n = 455)		
	Beta	**SE**	**p-value**	**Beta**	**SE**	**p-value**
Intercept	**240.07**	**22.76**	**<.0001**	**212.38**	**25.90**	**<.0001**
Sexual orientation (Ref = CH)			0.58			0.08
MH or CHwSS	4.53	4.67	0.33	14.96	8.03	0.06
Bisexual,Lesbian, or Gay	−1.90	6.22	0.76	12.00	8.10	0.14
Hours sleep, previous night	**−7.37**	**1.63**	**<.0001**	−1.18	3.10	0.70
Hours sleep, typical night	−3.18	2.22	0.15	−4.94	2.64	0.06
Wake time today	**−0.0024**	**0.0003**	**<.0001**	**−0.0016**	**0.0005**	**<0.01**
Cigarette	**37.54**	**15.40**	**0.01**	1.89	12.31	0.88
Alcohol	3.60	3.64	0.32	−4.86	5.20	0.35
Perceived stress	3.79	2.17	0.08	5.45	4.09	0.18
Hormonal contraception use	−4.39	3.43	0.20			
Noncompliance (wakeup sample)	−6.19	7.84	0.43	4.84	30.07	0.87

Notes: Variables centered around their means are hours sleep in previous night, hours sleep on a typical night, waking time, perceived stress score. Sexual orientation categories: (1) Bisexual, Gay or Lesbian; (2) MH, CHwSS = Mostly Heterosexual or Completely Heterosexual with same-sex partners; (3) CH = Completely Heterosexual with no same-sex partners (Ref). All numbers are rounded to nearest zero.

Bold indicates statistical significance.