Ethnicity and Puberty As Potential Indicators of Diurnal Cortisol Rhythm Differences in Adolescents

Lindsey Rosenthal
PSY 320
Neuroscience Program
Smith College
Northampton, Massachusetts, USA

Number of figures: 5

Number of tables: 0

Number of pages: 13

Author Note: To contact the author, email lhrosenthal@smith.edu.

Key Words: adolescents, diurnal cortisol rhythm, ethnicity, puberty

Abstract

Differences in diurnal cortisol rhythms have been found between populations of racial/ethnic minorities and their caucasian counterparts in the United States, indicating that the psychosocial stressor of race- and ethnicity- based discrimination serves as a factor in HPA axis dysregulation. Published research has presented conflicting results between different sexes and age groups. In this study, I utilized publicly available data from van Dammen et al. (2020) and examined differences in daily cortisol output, diurnal cortisol slope, and cortisol awakening response between adolescents (M = 15.4 years old , SD = 2.07) of *Dutch* and *Other* minority ethnicities (n=83, 60.24% girls, 6% *Other Ethnicity*). Consistent with prior research, adolescents from the *Other ethnicity* group had blunted diurnal slopes almost half as steep as adolescents from the *Dutch ethnicity* (caucasian) group. Furthermore, in a comparison between peers of both ethnic groups within the same pubertal development stage, adolescents of the *Other ethnicity* group showed lower peaks in cortisol concentration. When adolescents were grouped by their stage in pubertal development, regardless of ethnicity, a cyclic-like wave pattern was observed in daily cortisol output, diurnal cortisol slope, and cortisol awakening response. Across all three measures, cortisol concentration peaked or leveled off close to the end of pubertal development. While statistical testing did not show significant differences between groups, due to uneven group distributions and a limited sample size, these findings indicate that ethnicity and pubertal development stage may serve as potential indicators of differences in diurnal cortisol rhythm measures of adolescents.

Research Question

How do ethnicity and puberty affect diurnal cortisol rhythms and levels in adolescents?

Background

The psychosocial stressor of race-based discriminatory experiences has been found to cause adverse changes in health status and increase behavioral health risks (Williams and Mohammed, 2013). Dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis can be due to stressful experience of race- or ethnicity-based discrimination or prejudice, and might serve as a potential explanation or factor in health disparities. Chronic stress is associated with dysregulation of the HPA axis that results in altered basal diurnal patterns of cortisol secretion by the adrenal cortex (Clow et al., 2010). Cortisol is a biological marker that can be measured in saliva. It is biologically active and reflects a circadian rhythm due to reliably responding to plasma cortisol frequent changes in concentration (El-Farhan et al., 2017).

Blunted diurnal cortisol rhythms, or rhythms that have a less steep daily slope and are less healthy, have recently been found in healthy adults who faced low-moderate childhood adversity or trauma (Kuras et al., 2017). This finding indicates that even low-to-moderate trauma or adversity in childhood can contribute to basal HPA dysregulation. Recent research on everyday perceived discrimination (racial, gender, age, height, or weight) over the course of 1 year during adolescence has indicated that increased frequency of discriminatory experiences is associated with higher daily cortisol levels, less steep diurnal slopes, and lower wake and bedtime cortisol levels (Huynh et al., 2016).

When compared to their white counterparts, individuals of different racial and ethnic identities have been found to have differences in HPA axis activity. A particular group of interest and importance are adolescents due to their unique period in neurodevelopment and their hyperawareness of social environment/status. A recent study found that American youth from racial/ethnic minority groups tend to show flatter diurnal cortisol slopes when compared to Whites and controlled for age and sex (Deer et al., 2018). There is limited data on the difference in magnitude of cortisol rhythm dysregulation for most racial/ethnic minority groups. The majority of published literature has only focused on the comparison between African Americans and Whites. Perceived racial discrimination is a potential cause of flatter diurnal cortisol slopes, differences in CARs, and average cortisol levels (Adam et al., 2015). African American adolescents and young adults also differ in cortisol rhythm patterns, making neurodevelopmental stages a potentially important factor in diurnal cortisol rhythms and patterns (Adam et al., 2015). Current research on other racial and ethnic groups is limited.

Stage in pubertal development may be a factor in explaining the differences of diurnal cortisol rhythms. Sex differences may also have an indirect effect on differences in cortisol rhythms as females tend to reach sexual maturity at a younger age then males. Using the Tanner stages to assess pubertal development stage, Netherton et al. (2004) assessed salivary cortisol levels of males and females in different stages of puberty. The study found that mid-postpubertal females had higher morning cortisol levels than males in equivalent pubertal stages. No differences were found between the sexes at pre-early pubertal stages, indicating that changes in the HPA that occur during puberty impact cortisol secretion.

Although associations between race-based discrimination and altered diurnal cortisol rhythms have been found, many studies have presented conflicting results between different sexes and age groups. Differences in cortisol rhythms and levels during adolescence could be linked to physiological changes associated with puberty. Additionally, the previously mentioned findings have been reported by an extremely limited number of studies, mostly done in the United States, making this an emerging topic of importance for understanding health disparities. In this report, I aim to analyze the effects that ethnic identity and pubertal stage have on diurnal cortisol rhythms in adolescents from another eurocentric/mostly caucasian society, the Netherlands, to see if these differences are observed in other countries. To my knowledge, studies focused on ethnicity/race and diurnal cortisol rhythms have not been conducted in the Netherlands.

Methods

Participants

Data was publicly available through PLOS ONE and drawn from the paper "Sex-specific associations between person and environment-related childhood adverse events and levels of cortisol and DHEA in adolescence" by van Dammen et al. (2020). The participants from this study were 74 boys and 116 girls, 12-18 years old with a mean age of 15.7 years (SD = 2.0). Participants were recruited from several schools in the Netherlands with permission from the school and their parents.

Demographic data of participants was collected including age, ethnicity, SES, and sex. SES scores were based on the relative Dutch average (0), ranging from -2.43 to 1.91. Each participant was assigned a Puberty Development Scale (PDS) score. These scores were based on a self-reported measure of physical development and pubertal status. There was a different set of questions for males and females. The responses were coded on a 4-point scale: 1 = no pubertal development and 4 = completed pubertal development (van Dammen et al., 2020). Participants were also assigned a childhood adversity score based on the self-reported Adverse Life Events Questionnaire (ALEQ) where they were asked to indicate which type of adverse event, including

both person- and environment-related, they have experienced, how many times it had occurred and at what age it occurred.

A subset of participants were analyzed as the sample for this paper after removing participants with any incomplete entries (n=83, *M* = 15.4 years old, 60.24% girls). Based on parameters from the original paper, 78 participants were coded as *Dutch ethnicity* and 5 participants were coded as *Other ethnicities*. Countries of origin for individuals within the "*Other ethnicity*" group include China, Brazil, Somalia, and Morocco.

Procedure

To measure diurnal rhythms, salivary cortisol rhythms must be collected at times that coincide with important points in the diurnal rhythm. The first sample must be taken when an individual wakes up to obtain their initial morning level. The next time point is 30-minutes post wake-up, at which the rhythm should reach a sharp peak. This is followed by a decline throughout the day. Multiple samples, spaced a few hours apart, are normally obtained during this decline. Based on the Methods from van Dammen et al. (2020), students were given four test tubes for saliva collection on one day. Students were instructed to passively drool saliva into the tubes at four specific points during the day: right after waking up in the morning, 30 minutes after waking up while still in bed, at 12 noon, and a final sample at 8pm. They were told to not eat 30 minutes prior to saliva collection and to store samples in the freezer. Cortisol concentrations were obtained from salivary cortisol samples via immunoassay with time-resolved fluorescence endpoint detection.

Statistical analysis

The dataset was downloaded into R Studio for analysis using the readr, dplyr, ggplot, and tidyr packages. The variables analyzed were sex, ethnicity, age category, puberty development scale score (PDSscore), SES, AUCg, CAR, cortisol1, cortisol2, cortisol3, and cortisol4. Each cortisol# variable is representative of one of the four samples taken throughout the day in chronological order. The Mann-Whitney U test was used to compare AUC_g, diurnal slope, and CAR between participants of *Dutch ethnicity* and *Other ethnicity*.

Area under the curve (AUC_g), or total daily cortisol output, was calculated with 4 repeated measures and took the difference between the collection time intervals, in hours, into account (Prussner et al., 2003). The $t_2$interval was estimated by the authors from the original paper based on the mean wake-up time of 7:30am by study participants. Measures with respect from the ground (zero) were used. The following summation formula was used to compute AUC_g for each group.

$$AUC_g = \sum_{i=1}^{n-1} \frac{(m_{(i+1)} + m_i)^* t_i}{2}$$

$$AUC_g = \frac{(m2 + m1)^* t1}{2} + \frac{(m3 + m2)^* t2}{2} + \frac{(m4 + m3)^* t3}{2}$$

$$t_1 = 0.5,\ t_2 = 4,\ t_3 = 8$$

Following the methods from Deer et al. (2018) and Huynh et al. (2016), diurnal cortisol slopes were calculated by subtracting the last evening salivary cortisol sample concentration from the first salivary cortisol sample concentration, divided by the time in hours between the two samples.

$$Diurnal\ Slope = \frac{cortisol4 - cortisol1}{12.5\ hours}$$

Cortisol awakening response (CAR) is defined as the increase in cortisol concentration during the first 30 minutes after waking up, relative to wake up cortisol concentration (Ross et al., 2014). CAR was provided in the original data sample and was calculated by subtracting the waking cortisol sample concentration from the 30-minutes post waking cortisol sample concentration.

$$CAR = cortisol2 - cortisol1$$

Results

Average diurnal cortisol rhythms were charted for each group, *Dutch ethnicity* and *Other ethnicity*, and a visual comparison was done due to the difference in group sizes (Figure 1. & Figure 2.). A substantial visual difference is observed between Figure 1 and Figure 2 for samples collected at 7:30 am and 8 am. These time points were estimated by authors from the paper that the dataset was pulled from. At 7:30 am, the *Dutch ethnicity* group (n = 78) has an average cortisol concentration of $7.07\ nmol/l$ (SD = 3.722), while the *Other ethnicity* group (n = 5) has an average cortisol concentration of $4.59\ nmol/l$ (SD = 2.259). At 8 am, the *Dutch ethnicity* group (n = 78) has an average cortisol concentration of $10.11\ nmol/l$(SD = 4.366), while the *Other ethnicity* group (n = 5) has an average cortisol concentration of $7.31\ nmol/l$ (SD = 3.261). The difference in AUC_g between the two groups was $2.83\ nmol/l\ *\ hour$,with the *Dutch ethnicity* group having a higher total daily cortisol output (W = 224, $p = 0.58$). Both groups showed a negative diurnal slope, however the diurnal slope for the *Dutch ethnicity* group was almost twice as steep as the *Other ethnicity* group (W = 281, $p = 0.10$). Average CAR between the groups differed by $0.325\ nmol/l$ (W = 203, $p = 0.88$).

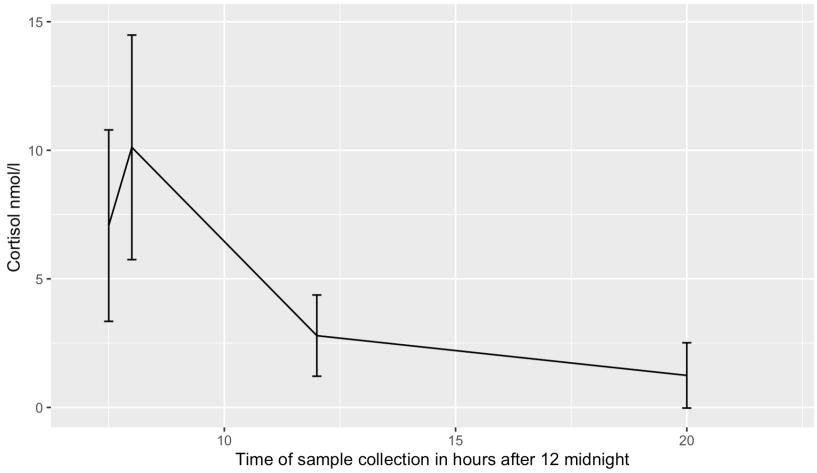

Figure 1. Average diurnal cortisol rhythm for *Dutch ethnicity* (n =78). Standard deviation bars are included for each sample collection time point. CAR = $3.045\ nmol/l$, $AUC_g = 46.27\ nmol/l * hour$,
$Average\ Diurnal\ Slope = -0.4659\ nmol/l/hour$.

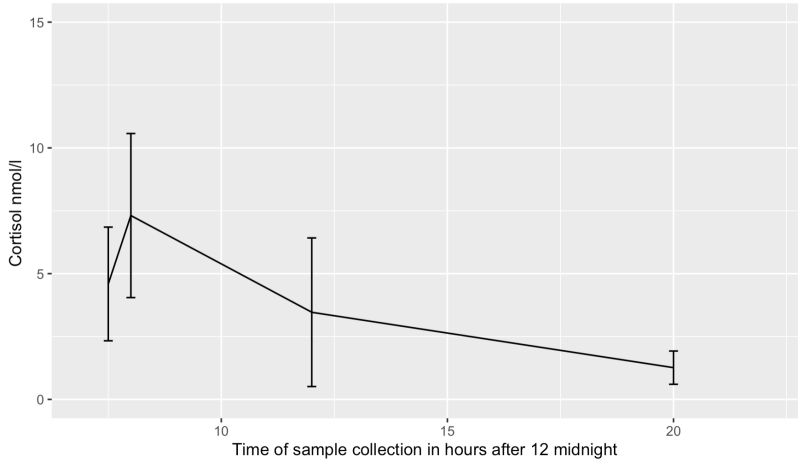

Figure 2. Average diurnal cortisol rhythm for *Other ethnicity* (n = 5). Standard deviation bars are included for each sample collection time point. CAR = $2.72\ nmol/l$, $AUC_g = 43.44\ nmol/l * hour$,
$Average\ Diurnal\ Slope = -0.26624\ nmol/l/hour$.

The individual diurnal cortisol rhythms of all five participants from the *Other ethnicity* group were graphed across the 12.5 hour collection time frame. Each participant had a different rhythm, and all followed the expected pattern of a peak in cortisol concentration for the second sample except one participant, shown as the lower blue line (Figure 3.). The participant with the PDS score of 1.4 had the highest 8 am peak cortisol concentration level at$10.69\ nmol/l$. The participant with the PDS score of 2.2 had the highest CAR at $4.54\ nmol/l$.The participant with

the PDS score of 3 (that has a higher peak cortisol concentration between the two participants with PDS score of 3) had the steepest diurnal slope of $-\ 0.5384\ nmol/l/hour$.

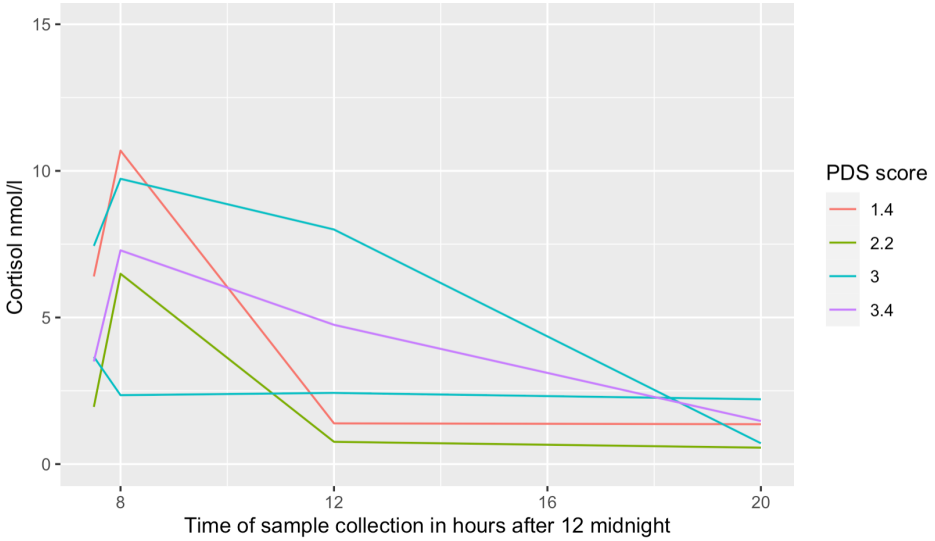

Figure 3. Individual diurnal cortisol rhythms for all 5 of the *Other ethnicity* study participants. Each participant is represented by a single line. The color of the line indicates the individual's PDS score as noted in the key.

Visible differences in diurnal rhythms, when grouped by PDS score, were seen when individual diurnal cortisol rhythms were charted across the 12.5 hour sample collection time frame (Figure 4.). A somewhat cyclical pattern can be observed: Lower peaks for PDS scores 1-1.4, a rise in peak cortisol concentrations for 1.6-2.4, a decrease at 2.6, another rise in peak cortisol concentrations for 2.8-3, another decrease at 3.2, and a final rise in peak cortisol concentrations 3.4-4. When compared to peers of the same pubertal development stage, differences are shown between participants of *Dutch* and *Other ethnicities* with individuals from the *Other ethnicity* group showing lower peaks in cortisol concentration, aside from the individual with a PDS score of 1.4.

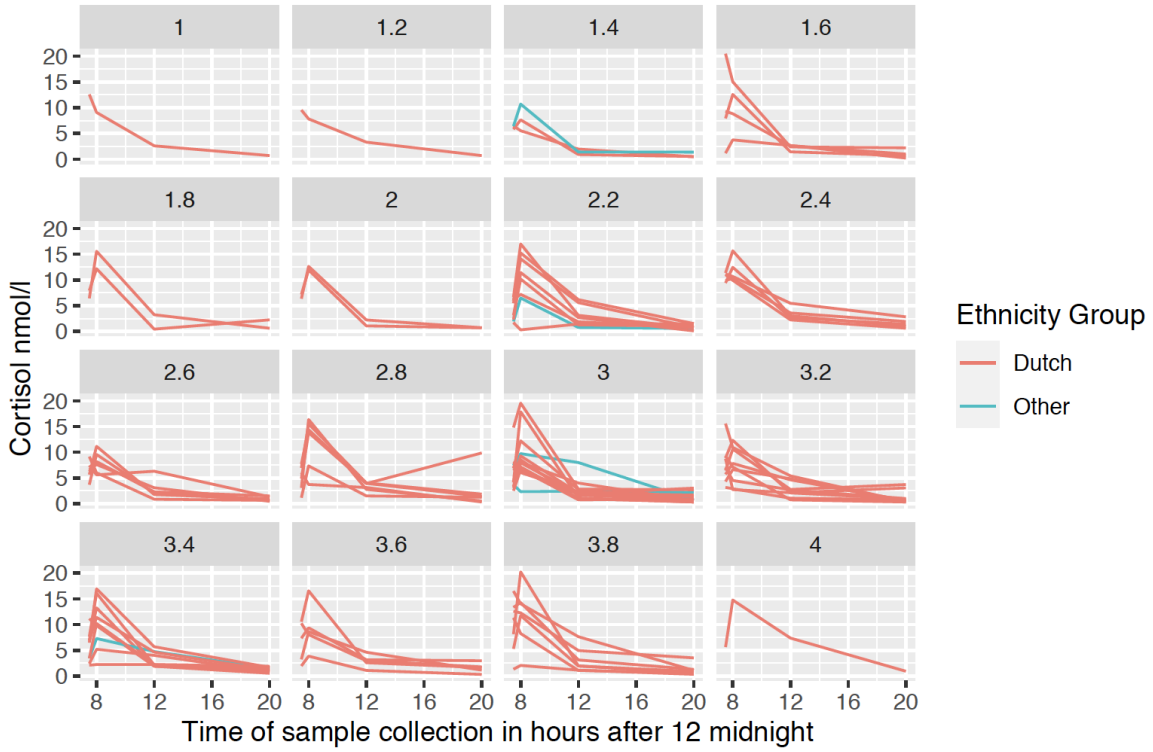

Figure 4. Individual diurnal rhythms grouped by PDS score (n = 83). Each participant is represented by a single line. The color indicates the ethnicity of the individual: red is *Dutch* and blue is *Other*.

Cyclic-like patterns and slight sex differences were seen in diurnal slope, AUC_g, and CAR. For diurnal slope, the valleys are seen at PDS scores of 2.2 and 2.8, while peaks are seen at 2.4 and 3.2 for both sexes, and an additional peak at 3.8 for girls (Figure 5A). For AUC_g, boys and girls showed different total daily output at the PDS score of 2.2, but then aligned to show a similar pattern with a valley at PSD scores 2.6-3 (Figure 5B). The peak in CAR for both sexes was at the PDS score of 2.2, while also showing a somewhat cyclic pattern with dips at 2.4 and 3.2 (Figure 5C).

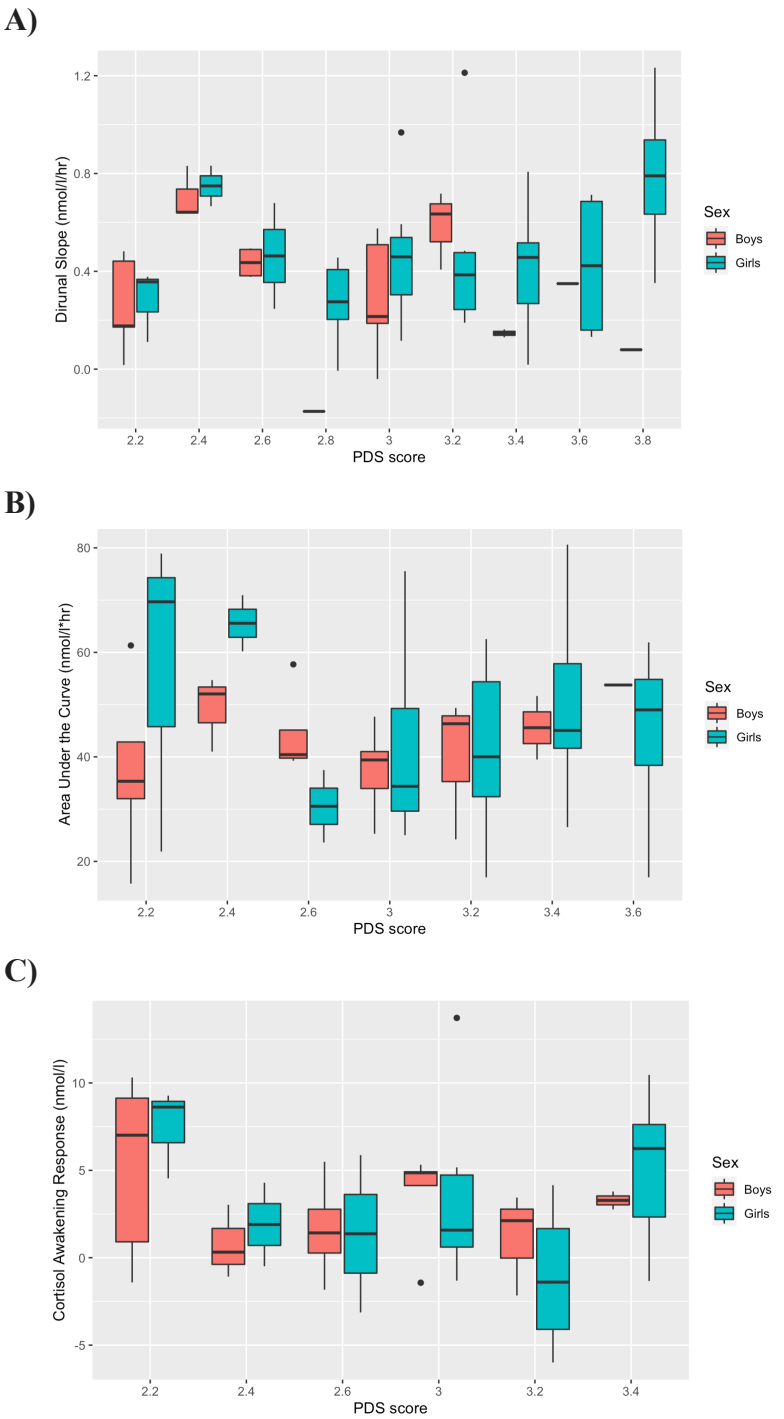

Figure 5. Box plots showing how cortisol rhythm markers vary by different PDS scores, grouped by sex (n=83). The color of each box indicates the sex of the group, as noted in the key. **(A)** Diurnal cortisol slopes of participants, grouped by PDS score and sex. **(B)** AUC_g of participants, grouped by PDS score and sex. **(C)** CAR of participants, grouped by PDS score and sex.

Discussion

My results indicate that the ethnicity of a child in adolescence likely impacts their diurnal cortisol rhythms and total daily output. Although the sample size of my Other ethnicity group was extremely small, an obvious visible difference was seen between the two groups. When the slopes of the two groups were calculated, it was found that the *Dutch ethnicity* group had a diurnal slope almost twice as steep as the *Other ethnicity* group. This finding reflects previously published studies that found adolescents from minority groups had blunted or flatter diurnal slopes when compared to their White counterparts (Adam et al., 2015; Deer et al., 2018). Perceived racial discrimination during adolescence has been linked to patterns of flatter diurnal slopes, lower waking cortisol levels, and lower daily cortisol output in Black adults (Adam et al., 2015). Consistent with previous research focused on race, I found that the average daily cortisol output and waking cortisol levels were lower in the *Other ethnicity* group than the *Dutch ethnicity* group, indicating that perceived everyday ethnicity-based discrimination could serve as a factor in HPA axis dysregulation. This, however, slightly varies from research on all types of perceived everyday discrimination in adolescents which has found that higher total daily cortisol outputs are associated with higher frequency of perceived everyday discrimination (Huynh et al., 2016). This could mean that chronic stress and trauma directly related to race- or ethnicity-based discrimination affects the HPA axis differently.

Previous research has shown sex specific differences in cortisol levels of adolescents (Netherton et al., 2004; van Dammen et al., 2020). When studying adolescents of the same age or age group, the females and males are likely in different stages of puberty. Based on my results, pubertal stage may be associated with HPA axis activity and also serve as a factor for cortisol differences seen between the sexes. This mirrors published literature that confirms the puberty-HPA stress hypothesis; that stress responses of the HPA axis increase as sexual maturity is reached (Gunnar, et al., 2009). A cyclic-like wave pattern was observed in my analysis when diurnal slope, total daily cortisol output, and CAR of both sexes were charted by PDS score. I was not able to test for the significance of PDS score since I had 16 different groups, of which many were missing participants or had a small number of participants. Previous studies have conflicting findings. The paper that I got the dataset from found that PDS score was not associated with results collected from salivary cortisol samples and AUC_g cortisol levels (van Dammen et al., 2020). However, Netherton et al. (2004) used the Tanner scale for determining pubertal development and found that morning cortisol levels were significantly higher in mid-post puberty females than males. While I looked at CAR and used the Pubertal Development Scale, my results show a similar trend of females with a much higher median CAR than males with a PDS score of 3.4, which is considered almost fully mature.

These findings only serve to indicate ethnicity and pubertal development stage as potential indicators of differences in diurnal cortisol rhythm measures due to multiple limitations. This

report was limited to publicly available datasets online. This dataset was obtained from a study conducted in the Netherlands, while most of the background research was conducted in the United States. This is due to the fact that I was unable to find any published research on the effect of race/ethnicity on cortisol rhythms from the Netherlands. The Netherlands and the United States do have similarities, as both countries are majority caucasian. In a report published by the Netherland's Social and Cultural Planning Office from 2019, it was found that citizens of Moroccan or Turkish descent, and Muslims encounter a "great deal" of unequal treatment, negative attitudes, and are perceived as threatening (Ministerie van Volksgezondheid, Welzijn en Sport, 2020). This is similar to how many ethnic and racial groups in the United States are treated or perceived. However, the countries differ greatly in health care accessibility and affordability. The Netherlands uses a universal social health insurance approach where the financing is mostly public and costs are monitored by the government (Tikkanen et al., 2020a). This is different from the healthcare approach in the United States which does not have a universal healthcare coverage program (Tikkanen et al., 2020b). A country's health insurance program may also impact SES distribution. In the United States, there is an extremely wide range in household income. In 2019, the household income ratio between the top 95th percentile and 50th percentile was 3.93, meaning households in the top 5% made almost four times the national median income (US Census Bureau, 2020). There are also huge racial wealth gaps in America. In a 2019 survey, it was revealed that Black and Hispanic households in the United States have a mean and median wealth less than 20% of White household's wealth (Bhutta et al., 2020). The dataset used in this study measured SES based on the Dutch average neighborhood social status denoted at zero (0) with a range of -2.34 to 1.91. This wealth gap in the dataset from the Netherlands is not completely representative of or as substantial as the wealth gap seen in the United States, but it is still present. The SES values for participants in my sample's Other ethnicity group ranged from -0.3803 to 0.6618, which follows a similar and less-drastic trend of that seen in the United States.

The salivary cortisol samples were only taken on 1 single day, leading to potential collection errors and incorrect assayed concentration values. This is likely the case for participants that had peak values as their first wake-up cortisol sample, since the normal diurnal pattern shows a peak at the second post-wake up cortisol sample. In the future, by collecting samples across multiple days, an average for each individual could be calculated and then used for a more accurate analysis..

Although my results were not statistically significant, there were many limitations with the dataset. After cleaning the data, it ended up including a small sample size of 83 participants which was less than half of the original dataset. The ethnic groups also differed greatly: 78 to 5. Non-linear patterns of CAR, diurnal slope, and AUC_g likely affected the significance of the p-values when testing for a relationship with ethnicity. There are also many other variables to consider like SES, sex, PDS score, ALEQ score, and age that may impact cortisol rhythm

measures that I was unable to control or adjust for, making it difficult to to analyze the effects of individual factors. In the future, I would increase the sample size and have more even distributions between groups.

Knowing that altered diurnal cortisol rhythms have negative implications on health, and that individuals from racial and ethnic minority groups are more at risk for HPA axis dysregulation due to higher rates of chronic stress and trauma from everyday discrimination, finding effective methods for re-regulation of the HPA axis is extremely important. Re-regulation of cortisol rhythms and levels via behavioral intervention could positively impact overall health, reduce risk of disease, and decrease prolonged effects of mental health disorders. A limited number of studies have been done on regulating diurnal cortisol rhythms of infants and young children, but of the published studies, psychosocial interventions that target children and parents do work (Slopen et al., 2014). In a recent 3 year follow-up study on a racially diverse sample of Child Protective Services-referred infants -- now preschool aged children, previously blunted diurnal cortisol rhythms were successfully regulated to show more "normal" rhythms after completing a 10 week Attachment and Biobehavioral Catch-up (ABC) parent intervention program (Bernard et al., 2015). This ABC program was designed for parents of young children who are at risk for parental neglect and may not be a viable method for regulation in adolescents, but shows that re-regulation of the HPA axis is possible and potentially long lasting. Future research on re-regulation via psychosocial intervention during adolescence should be conducted. The impact of factors like race/ethnicity and stage in pubertal development need to be considered when determining the effectiveness and timing of the interventions.

Acknowledgments

I would like to thank Professor Mary Harrington and Osman Keshawarz for their guidance and assistance in the data searching and analyzing processes. I would also like to thank my peers in PSY 320 for the support and encouragement throughout my first fully remote Zoom semester.

References

Adam, E. K., Heissel, J. A., Zeiders, K. H., Richeson, J. A., Ross, E. C., Ehrlich, K. B., … Eccles, J. S. (2015, August 28). *Developmental histories of perceived racial discrimination and diurnal cortisol profiles in adulthood: A 20-year prospective study*. Psychoneuroendocrinology. https://www.sciencedirect.com/science/article/pii/S0306453015008914

Bernard, K., Hostinar, C. E., & Dozier, M. (2015, February). Intervention Effects on Diurnal Cortisol Rhythms of Child Protective Services-Referred Infants in Early Childhood: Preschool Follow-up Results of a Randomized Clinical Trial. *JAMA Pediatrics, 196(2)*, 112-119. https://jamanetwork.com/journals/jamapediatrics/fullarticle/1939057

Bhutta, N., Chang, A. C., Dettling, L. J., Hsu, J. W., & Hewitt, J. (2020, September 28). Disparities in wealth by race and ethnicity in the 2019 survey of consumer finances. Retrieved May 12, 2021, from https://www.federalreserve.gov/econres/notes/feds-notes/disparities-in-wealth-by-race-and-ethnicity-in-the-2019-survey-of-consumer-finances-20200928.htm

Clow, A., Hucklebridge, F., & Thorn, L. (2010). The cortisol awakening response in context. *International review of neurobiology*, *93*, 153–175.

Deer, L. K., Shields, G. S., Ivory, S. L., Hostinar, C. E., & Telzer, E. H. (2018, December). Racial/ethnic disparities in cortisol diurnal patterns and affect in adolescence. *Development and psychopathology, 30(5),* 1977-1993. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6345388/

El-Farhan, N., Rees, D. A., & Evans, C. (2017). Measuring cortisol in serum, urine and saliva – are our assays good enough? *SAGE Journals.* https://journals.sagepub.com/doi/10.1177/0004563216687335?url_ver=Z39.88-2003&rfr_id=ori%3Arid%3Acrossref.org&rfr_dat=cr_pub++0pubmed

Gunnar, M. R., Wewerka, S., Frenn, K., Long, J. D., & Griggs, C. (2009). Developmental changes in hypothalamus-pituitary-adrenal activity over the transition to adolescence: normative changes and associations with puberty. *Development and psychopathology*, *21*(1), 69–85. https://doi.org/10.1017/S0954579409000054

Huynh, V. W., Guan, S. A., Almeida, D. M., McCreath, H., & Fuligni, A. J. (2016). Everyday discrimination and diurnal cortisol during adolescence. *Hormones and behavior*, *80*, 76–81. https://doi.org/10.1016/j.yhbeh.2016.01.009

Kuras, Y. I., Assaf, N., Thoma, M. V., Gianferante, D., Hanlin, L., Chen, X., Fiksdal, A., & Rohleder, N. (2017). Blunted Diurnal Cortisol Activity in Healthy Adults with Childhood Adversity. *Frontiers in human neuroscience*, *11*, 574. https://doi.org/10.3389/fnhum.2017.00574

Ministerie van Volksgezondheid, Welzijn en Sport. (2020, April 02). Perceived discrimination in the Netherlands 2019. Retrieved May 19, 2021, from https://www.scp.nl/publicaties/publicaties/2020/04/02/ervaren-discriminatie-in-nederland-ii

Netherton, C., Goodyer, I., Tamplin, A., & Herbert, J. (2003). Salivary cortisol and dehydroepiandrosterone in relation to puberty and gender. *Psychoneuroendocrinology, 29(2)*, 125-140. https://www.sciencedirect.com/science/article/pii/S0306453002001506?via%3Dihub

Pruessner, J. C., Kirschbaum, C., Meinlschmid, G., & Hellhammer, D. H. (2003). Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change. *Psychoneuroendocrinology*, *28*(7), 916–931. https://doi.org/10.1016/s0306-4530(02)00108-7

Ross, K. M., Murphy, M., Adam, E. K., Chen, E., & Miller, G. E. (2014). How stable are diurnal cortisol activity indices in healthy individuals? Evidence from three multi-wave studies. *Psychoneuroendocrinology*, *39*, 184–193. https://doi.org/10.1016/j.psyneuen.2013.09.016

Slopen, N., McLaughlin, K. A., & Shonkoff, J. P. (2014). Interventions to improve cortisol regulation in children: a systematic review. *Pediatrics*, *133*(2), 312–326. https://doi.org/10.1542/peds.2013-1632

Tikkanen, R., Osborn, R., Mossialos, E., Djordjevic, A., & Wharton, G. (2020, June 5). International Health Care System Profiles: Netherlands. https://www.commonwealthfund.org/international-health-policy-center/countries/netherlands

Tikkanen, R., Osborn, R., Mossialos, E., Djordjevic, A., & Wharton, G. (2020, June 5). International Health Care System Profiles: Netherlands. https://www.commonwealthfund.org/international-health-policy-center/countries/united-states

US Census Bureau. (2020, September 15). Income and poverty in the United States: 2019. https://www.census.gov/library/publications/2020/demo/p60-270.html

van Dammen, L., de Rooij, S. R., Behnsen, P. M., & Huizink, A. C. (2020). Sex-specific associations between person and environment-related childhood adverse events and levels of cortisol and DHEA in adolescence. *PLOS ONE*, *15*(6). https://doi.org/10.1371/journal.pone.0233718

Williams, D. R., & Mohammed, S. A. (2013). Racism and Health I: Pathways and Scientific Evidence. *American Behavioral Scientist*, *57*(8), 1152–1173. https://doi.org/10.1177/0002764213487340