The Link between Hair Cortisol Levels and Physical Activity in a Study of Young Adolescents

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Abstract

This study explored associations between different levels of physical activity and cortisol levels in hair among adolescents. Although research has found that physical activity can be beneficial for reducing illness and stress in populations, studies have found a correlation between strenuous physical activity and elevated salivary and hair cortisol levels. This study employs secondary analysis of data from a representative sub-sample of 534 adolescents aged 11 to 17 years who participated in two linked NIH funded studies on the health and well-being of urban adolescents. Through an in-home survey, youth were asked three questions on the intensity and volume of physical activity from the previous week, rating from mild to strenuous physical activity, and the number of days the activity was done. The three variables – mild, moderate and strenuous activity - were analyzed via regression analyses as continuous measures to assess linear associations. No significant linear relationships were found with this analysis, therefore the recommended reference range of exercising 6-7 times a week was compared to exercise categories based on frequency of activity (never, 1-3 times weekly, 4-5 times weekly, and 8 or more times weekly) . The results found that hair cortisol levels were significantly higher in adolescents who strenuously exercised eight times or more a week in comparison to those who strenuously exercised 6-7 hours per week as recommended($p<0.05$). For moderate physical activity, all frequencies of exercise had marginally or significantly higher hair cortisol levels than those who exercised six to seven times a week. No relationships were found between hair cortisol and mild physical activity. The data suggest that strenuous or moderate physical activity over eight times a week was correlated with higher hair cortisol levels compared to the recommended amount of daily exercise. In addition, the data found that those who do not engage in any moderate physical activity had higher hair cortisol levels compared to the reference range.

This suggests that hair cortisol might be an important biomarker in the research of studying the effects of physical activity on physiological measures of stress.

Keywords: cortisol, adolescents, physical activity, stress

Introduction

Exercise and activity play a large role for physiological and psychological well-being in adolescents. According to the US Department of Health and Human Services (USDHHS), children and adolescents are recommended to engage in at least 60 minutes or more of physical activity per day (2018). It is recommended that aerobic activity should comprise most of the activity and muscle strengthening activities should occur at least three days per week (Hill et al., 2008). Studies have shown that physical activity benefits cognition and metacognition including inhibition, working memory, and academic behavior (Álvarez-Bueno et al., 2017; Lees et al., 2013). Enriched physical activity programs with cognitive tasks help improve inhibitory skills and executive function (Álvarez-Bueno et al., 2017). There are no studies showing that aerobic exercise has a negative impact on cognitive and psychosocial function (Lees et al., 2013). In addition, regular, moderate exercise is associated with a lower risk of infection such as respiratory viral infections when compared to a completely sedentary state (Gleeson, 2007; Martin et al., 2009).

In addition to physiological well-being, cross sectional and longitudinal studies have shown that aerobic exercise training has antidepressant effects, can relieve anxiety, and can protect against harmful effects of stress (Salmon, 2001). Depression manifests with imbalances of serotonin, dopamine, noradrenaline and glutamate. In those that did aerobic exercise, levels of serotonin increased, showing similar effects of antidepressants (Mikkelsen et al., 2017). A study of 147 adolescents showed that those who reported greater physical activity also reported less stress and lower levels of depression (Norris et al., 1992). In this study, adolescents were separated into three different levels of exercise and those who had high intensity exercise reported significantly less perceived stress (1992). Although this an older study, more current

research supports the findings that exercise can make an impact on mental health. Mikkelsen (2017) studied the relationship between exercise and endorphins. Endorphins play a role in helping the body endure pain and prolonged pain that are experienced during exercise (2017). When these are released, it can lead to a feeling of well-being (2017). This feeling in runners is commonly known as a runner's high. This demonstrates that exercise plays an important role on improving mood and wellness in the human body.

Exercise also can lessen the hypothalamic-pituitary-adrenal (HPA) axis response to stress. The HPA axis is involved in the production of cortisol. Cortisol is a catabolic biomarker that helps use glycogen and free fatty acids and levels rise after hard exercise (Budgett, 1990). Those with depression and anxiety have a hyperactive HPA axis and routine exercise has shown to provide more resilience for affected individuals when they are exposed to stressful situations (Mikkelson et al., 2017). Exercise has also shown to have many more benefits, including helping to prevent distracting or ruminating thoughts, increase feelings of accomplishment, and decrease inflammation (Mikkelson et al., 2017).

However, the amount, type, duration, and volume of exercise can also negatively impact levels of stress within the body. Cortisol, a stress hormone, is secreted by the adrenal cortex in response to physical, psychological, or physiological stressors (Hill et al., 2008). Cortisol is also released in response to physical exercise and plays an important role during exercise because of its catabolic, anti-inflammatory characteristics, and helps maintain blood pressure and plasma (Gerber et al., 2012). Exercise can act as a stimulus, resulting in significant increases in cortisol depending on the intensity and duration of the activity (Hill et al., 2008). For example, one study examined the influence of exercise intensity on the cortisol response after 30 minutes of exercise; the intensity of exercise was measured at levels of 40, 60, and 80% of $VO_{2\ max}$ (2008). They drew

plasma levels and found that cortisol was not released at 40% VO_{2max}, and that the most cortisol was released before and after the workout at 80% VO_{2max} (2008). These results implicate the specific intensities of exercise on cortisol release.

Excessive amount of exercise can contribute to overtraining syndrome and higher cortisol level. There is evidence that cortisol levels in addition to others in excessive training or too much physical activity can contribute to overtraining syndrome (Budgett, 1990).

There is a circadian rhythm of adrenal cortisol activity that is important to the balance of the immune system and it is sensitive to both physical and psychological stress that could be induced with excessive amounts of exercise (2001). Long-term endurance training has been shown to produce immunological changes that are associated with increased cortisol. (Clow et al., 2001). For example, post-exercise immune function is most depressed when exercise is intense and prolonged without food intake and periods of intensified training have shown to chronically suppress immune function (Gleeson, 2006). In addition, evidence exists that increased cortisol levels may predispose individuals to several diseases, depression (Skoluda et al., 2012), possibly myocardial infarction (Pereg et al., 2011), and when it is produced over sustained periods of time, it can contribute to cognitive impairment (Clow et al., 2001).

Cortisol levels have been measured with different methods such as plasma or saliva, but a new sampling method using hair samples can be used to measure longer-term cortisol levels in a population. Measuring acute cortisol levels using plasma or saliva can be unreliable due to changes in cortisol from food intake, smoking, physical activity, and the natural circadian rhythm (Gerber et al., 2012). Hair cortisol is unique, non-invasive, and captures the long-term mean cortisol secretion (Gerber et al., 2012). It doesn't decompose and is easy to collect and store

(2012). Hair cortisol concentrations have also been used to evaluate disturbances of the HPA-axis, which is a vital component of regulating the body's response to stress. (Wester et al., 2015).

In a study by Skoulda et al. (2012), the relationship between endurance sports and hair cortisol concentrations was investigated in a sample of 304 participants who were amateur endurance athletes compared to 70 controls. Athlete subgroups included 10km runners, half marathoners, marathoners, triathletes and cyclists (2012). Small hair strands were taken, and the results showed that hair cortisol concentrations were higher in the endurance athletes compared to the controls (2012). This demonstrates the need to further investigate the relationship between the frequency and intensity of exercise that produces the optimum amount of cortisol levels circulating within the body. The aim of this study is to describe and explore the relationship between the frequency and intensity of physical activity among adolescents and hair cortisol levels to achieve a better understanding of the optimum levels of exercise for developing adolescents.

Methods

Study Design

This is a secondary analysis of cross sectional data to describe and explore the associations between the frequency and intensity of physical activity and hair cortisol levels in adolescents. The data for this study were derived from two NIH funded studies. The first study was The *Adolescent Health and Development in Context (AHDC)* study (Browning, PI, 1R01DA032371), which is a prospective cohort study that looks at the impact of activity space exposures on the behavioral and health outcomes of a sample of adolescents aged 11-17 years in Franklin County, Ohio. The second study was The *Linking Biological and Social Pathways to Adolescent Health*

and Wellbeing study (Ford, PI, 1R21DA034960) in which a subsample of youth who participated in the first wave of the AHDC study had biomarkers of chronic stress collected for linkage to the AHDC study (Ford, et al., 2016). The studies are approved by The Ohio State University Institutional Review Board.

Sample

The sample included a representative subsample of 534 adolescents aged 11-17 years from urban Columbus, Ohio and the surrounding suburbs, Out of 614 participants, 534 were included in the final analytic sample as 74 adolescents were missing responses on physical activity and 5 were dropped due to outlying cortisol values.

Data Collection

The first wave of the AHDC study was analyzed for this study utilizing data from an in-home survey of the adolescent as well as cortisol levels assayed from the collection of their hair. Youth and caregivers who refused participation and youth with insufficient hair length (< 1 cm) were categorized as nonparticipants in the hair collection. Adolescent assent and parental permission for participation were obtained prior to data collection. To collect the hair, trained interviewers cut approximately 10-50 mg of hair from the posterior vertex region of the youth's scalp using a new pair of thinning shears. Adolescents were compensated $20 for their participation in the hair collection. Of the eligible sample, 91.3% of the youth provided a hair sample for cortisol- of these approximately 3.5% (n=18) had insufficient hair to cut (<1 cm) and 5.2% (n=27) refused participation in the biomarker collection overall (Ford, Boch, & McCarthy, 2016).

Measures

Independent Variable

The primary independent variables of interest in this study were the frequency of mild, moderate and strenuous physical activity

Three questions were asked related to the level of physical activity: "During a typical week (7 days), how many times on average do you do the following kinds of activities for more than 15 minutes during your free time? (1) Strenuous physical activity: exercise where your heart beats rapidly such as running, jogging, basketball, cheerleading, vigorous cycling, rollerblading, soccer, martial arts, aerobics, etc." (2) Moderate physical activity: exercise (exercise that is not exhausting), such as fast walking, easy bicycling, volleyball, easy swimming, etc." (3) Mild physical activity: mild exercise, such as yoga, bowling, golf, easy walking. Response options included: 0 times per week, 1 time, 2 or 3, 4 or 5, 6 or 7, or 8 or more times per week. For the purposes of this study, strenuous, moderate and mild physical activity was measured in the analysis as continuous measures and also categorical measures were created in which the US Department of Health and Human Services (USDHHS) recommended reference range of exercising 6-7 times a week (2018) was compared to exercise categories based on the following frequencies of activity: never, 1-3 times weekly, 4-5 times weekly, and 8 or more times weekly.

Dependent Variable

The dependent variable in this study was the level of cortisol in hair. The hair is assayed for the mean cortisol value at the Ford lab using adapted protocol by D'Anna-Hernandez et al. (2011) and Meyer et al. (2014). The first 1 cm to 3 cm of hair most proximal to the scalp was analyzed for cortisol, thus hair samples longer than 3 cm. were cut to that length. The hair was

then washed twice with isopropanol and dried over 1-3 days. The hair was then placed into a microcentrifuge tube, minced, and ground in Retsch 400 Mill. 1.1 ml of HPLC-grade methanol is added to the ground sample, incubated for 18-24 hours at room temperature. Samples were centrifuged at 5000g for 5 min at room temperature to pellet the powdered hair. The entire amount was then transferred to a clean microcentrifuge tube and the methanol removed through evaporation using a stream of air for 6-8 hours at room temperature. The cortisol extract was reconstituted in 100ul of Salimetric immunoassay cortisol analysis diluent buffer. Samples were assayed in duplicate and hair cortisol levels expressed as pg. /mg. Hair cortisol levels were logged for analysis to normalize the distribution.

Data Analysis Strategy

The analysis consisted of descriptive and multivariable linear regression analyses in which the latter controlled for hair length and weight only. The descriptive analyses examined the frequencies of the differing intensities of physical activity and the mean of hair cortisol. Multivariable linear regression analyses were conducted to examine associations between continuous and categorical measures of strenuous, moderate and mild physical activity and hair cortisol levels. Data analysis was conducting using SAS Software, Version 9.4 (Carey, NC) and the level of statistical significance was $p<0.05$.

Results

In total, 534 adolescents were included in the study sample. Table 1 shown below presents the demographic characteristics of the sample.

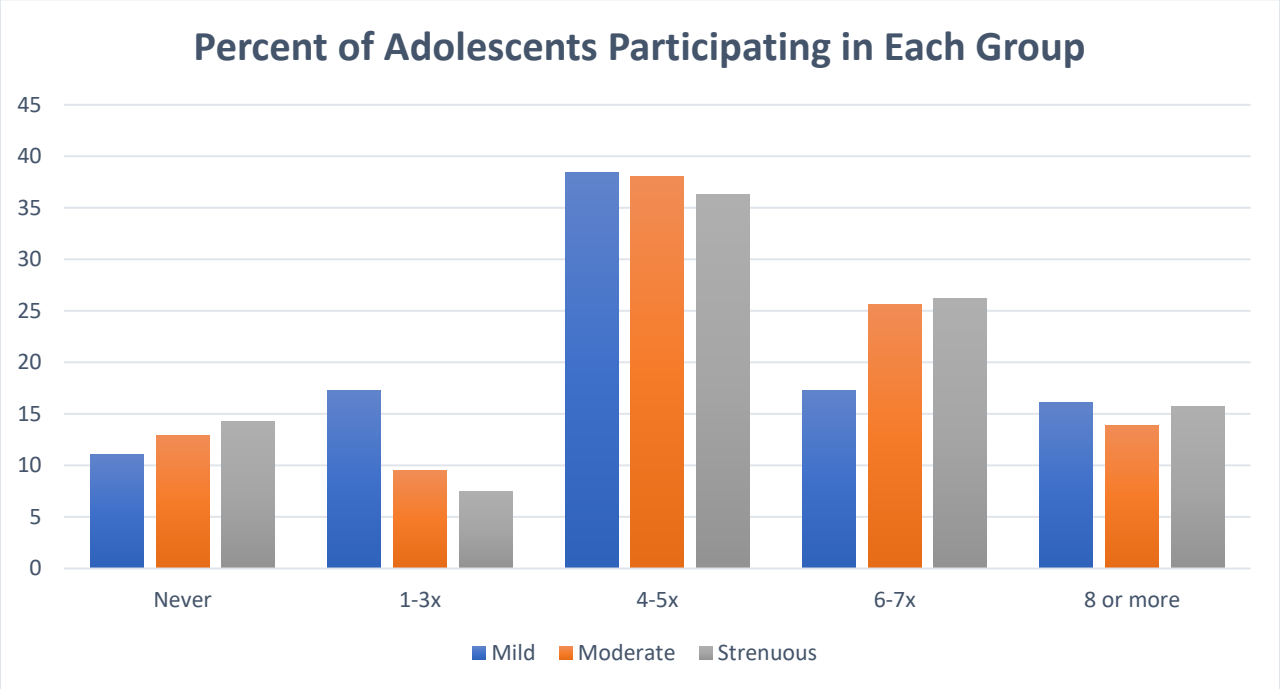
Table 1. Characteristics of the sample N=534

	n	**%**
Sex		
Male	275	51.50
Female	259	48.50
Race/ethnicity		
Black/African American	155	29.00
Multiracial	35	6.60
"other"	41	7.70
White	303	56.70
Primary caregiver level of education		
High school degree or less	90	16.90
Some college	162	30.30
Bachelor degree	159	29.80
Master degree or more	115	21.50
Missing response	8	1.50
	Mean	**(sd)**
Age (range 11-17)	14.70	(1.77)
Hair cortisol level in pg/mg (range 0.05 – 79.0)	5.10	(9.17)

Of the 534 adolescents, 51.50% were male and the average age was 14.70 years (range 11 to 17 years). With respect to race and ethnicity, 56.70% of the adolescents were categorized as non-Hispanic white, 29.00% were categorized as non-Hispanic black/African American, 7.70% were categorized as 'other' (due to small sample sizes, adolescents who self-identified as other, Hispanic or Asian were categorized as other) and 6.60% were categorized as multiracial. For the primary caregiver level of education, 16.90% of the adolescents had a primary caregiver with a

high school degree or less, 30.30% had a primary caregiver with some college, 29.80% had a primary caregiver with a bachelor's degree, 21.50% had a primary caregiver with a master's degree and 1.50% of the primary caregivers were missing on the response. Last, the mean hair cortisol level for the sample was 5.10 pg/mg with a range of 0.05 pg/mg to 79.00 pg/mg.

Figure 1 shown below depicts the frequency in which adolescents participated in strenuous, moderate and mild physical activity. Specifically, for strenuous physical activity, 76 adolescents (14.23%) reported they never exercised on average, 40 (7.49%) reported they exercised 1-3 times a week on average, 194 (36.33%) reported they exercised 4-5 times a week on average, 140 (26.22%) reported they exercised 6-7 times on average, and 84 (15.73%) reported they exercised eight or more times a week on average. For moderate physical activity, 69 adolescents (12.92%) reported they never exercised on average, 51 (9.55%) reported they exercised 1-3 times per week on average, 206 (38.01%) reported they exercised 4-5 times per week on average, 137 (25.66%) reported they exercised 6-7 times a week on average, and 74 (13.86%) reported they exercised eight or more times a week on average. For the mild physical activity, 59 adolescents (11.05%) reported they never exercised on average, 92 (17.23%) reported they exercised 1-3 times a week on average, 205 (38.39%) reported they exercised 4-5 times a week on average, 92 (17.23%) reported they exercised 6-7 times a week on average, and 86 (16.10%) reported they exercised eight or more times a week on average. With respect to the cortisol levels, the average hair cortisol level in the sample was 5.1 pg/mg, the standard deviation was 9.17 pg/mg, and the range was 0.02 pg/mg – 79.0 pg/mg



Multivariable Results

Multivariable linear analyses found no significant relationships between the continuous measures of strenuous, moderate or mild physical activity and adolescent hair cortisol levels (results not depicted in the Table). Significant results were found for the associations between the categorical measures of physical activity and hair cortisol levels and are presented the table shown below in Figure 1.

For strenuous physical activity, adolescents who reported they exercised eight or more times a week on average were found to have significantly higher hair cortisol levels in comparison to adolescents who reported they exercised within the reference range of 6-7 times per week. ($b=0.35$, $p<0.05$). No significant differences in hair cortisol levels were found between adolescents who reported lower levels of strenuous physical activity on average in comparison to the reference of 6-7 times a week on average. For moderate physical activity, adolescents who reported they never exercised ($b=0.46$, $p<0.05$) or that they exercised 8 or more

times a week on average (b=0.44, p<0.05) were found to have significantly higher hair cortisol levels than those who exercised in the reference range. In addition, adolescents who reported they engaged in moderate physical activity 1-3 times a week on average (b=0.26, p<0.10) or 4-5 times a week on average (b=0.26, p<0.10) were found to have marginally significant higher hair cortisol levels compared to those who exercised 6-7 times a week. For mild physical activity, there were no statistically significant differences in hair cortisol levels for youth who exercised 6-7 times a week on average (the reference) in comparison to those who engaged in less or more mild exercise.

Multivariable Regression Analysis of the Relationships between Physical Activity and Hair Cortisol Levels	**b**
Frequency of Strenuous Activity in a Typical Week (# Days)	
0	0.25
1-3	-0.03
4-5	-0.06
6-7 (reference)	------
8 or more	0.35*
Frequency of Moderate Activity in Typical week (# Days)	
0	0.46*
1-3	0.26~
4-5	0.26~
6-7 (reference)	------
8 or more	0.44*
Frequency of Mild Activity in Typical Week	
0	0.46*
1-3	0.06
4-5	0.01
6-7 (reference)	------
8 or more	-0.05
Controlling for hair length and hair weight ****p<0.05; ~p<0.10***	

Discussion

This study's purpose was to analyze how different frequencies of strenuous, moderate and mild physical activity were associated with hair cortisol levels in adolescents. In summary, the results suggest that higher levels of strenuous or moderate exercise (above the reference range) or no moderate exercise at all correlated with higher cortisol levels in comparison to adolescents who exercised in the recommended range of 6-7 times a week. Previous studies have suggested that different amounts of exercise can significantly impact cortisol levels (Hill, 2008; Skoluda, 2012) and this could impact mood and cognitive function (Clow et al., 2001). Although physical activity can have health benefits (Álvarez-Bueno et al., 2017; Lees et al., 2013), prolonged strenuous activity can negatively impact immune function and increase risk to infection (Nieman, 2003). This is shown in this study in which hair cortisol levels were significantly higher in those who engaged in strenuous or moderate physical activity eight or more times a week in comparison to those who exercised only 6-7 times per week.

The findings of this study showed no correlation between the level of mild physical activity and hair cortisol levels, which is an interesting finding as a recent meta-analysis found lower levels of salivary cortisol with yoga (Pascoe, Thompson, & Ski, 2017). One potential explanation for these discrepant findings may be the measurement of mild physical activity for this study in which differing types of activities were grouped together (e.g. slow walking, bowling, yoga) and perhaps not all types of activity have an effect on cortisol levels. This suggests that future studies could explore how specific forms of mild physical activity could impact cortisol stress levels.

Cortisol is not the only factor that plays a part in exercise and stress in adolescents. In previous studies, testosterone was shown to also have an impact on stress and overtraining within the body. The ratio of testosterone/cortisol has an impact on determining whether an athlete is effectively recovering from training or if their metabolism continues to break down (Budgett**,** 1990). This ratio between these two has been suggested for use as a marker of the status of protein turnover in an athlete (Petibois et al., 2002). If this ratio drops 30% below the basal values of an athlete before training, it might indicate that the athlete is training too intensely (2002). Budgett explains that intense, prolonged training will raise cortisol levels, but testosterone levels will become suppressed in men possibly due to the increased cortisol levels (1990). This shows that there could be more factors that play into the appropriate level of activity that produces the optimum amount of activity to engage in. Overtraining can correlate with a negative impact in immune function, mood, energy levels, and body regulation so testosterone could be another important factor to study in combination with cortisol to further explore the results of our study.

Several limitations to this study warrant discussion. First, this was an exploratory descriptive study that did not account for other stressors that could have been occurring in the youth's lives at the time such as home life, schoolwork, or social impacts that may have an effect on hair cortisol levels and also the frequency in which adolescents were able to engage in physical activity. Another limitation is that genetic factors were not taken into consideration which could alter the results in this study. In addition, some adolescents did not meet the criteria or were missing information required to be included in this study as out of 614 adolescents who had hair cortisol levels measured, only 534 were included in the analysis. Thus, it is possible the

results are biased due to the decrease in sample size. Future analyses with multiple imputation will be conducted, but they were beyond the scope of this current study.

Conclusion

Cortisol levels play an important role in stress, immune function, cognition and mood. Many things have an impact on the circulating cortisol levels in the body, and one important variable is physical activity. The CDC recommends exercising 6-7 times a week, but the findings of this study along with others have found that varying levels and frequencies of exercise can differentially impact cortisol levels. Most studies have examined cortisol levels in saliva, yet this only examines short-term cortisol levels. By studying hair cortisol levels, we can get a more robust picture of cortisol levels over time and may be able to better determine the optimal level of physical activity for adolescent health and development.

Works Cited

Álvarez-Bueno, C., Pesce, C., Cavero-Redondo, I., Sánchez-López, M., Martínez-Hortelano, J. A., & Martínez-Vizcaíno, V. (2017). The Effect of Physical Activity Interventions on Children's Cognition and Metacognition: A Systematic Review and Meta-Analysis. *Journal of the American Academy of Child & Adolescent Psychiatry*,729-738. doi:10.1016/j.jaac.2017.06.012

Anderson, T., Haake, S., Lane, A. R., & Hackney, A. C. (2016). CHANGES IN RESTING SALIVARY TESTOSTERONE, CORTISOL AND INTERLEUKIN-6 AS BIOMARKERS OF OVERTRAINING. Baltic Journal of Sport & Health Sciences, 1(100), 2-7. Retrieved October 20, 2017.

Budgett, R. (1990). Overtraining syndrome. *British Journal of Sports Medicine, 24*(4), 231-236. doi:10.1136/bjsm.24.4.231

Clow, A., & Hucklebridge, F. (2001). The impact of psychological stress on immune function in the athletic population. *Exercise Immunology Review, 7,* 5-17.

D'Anna-Hernandez KL, Ross RG, Natvig CL, Laudenslager ML. Hair cortisol levels as a retrospective marker of hypothalamic-pituitary axis activity throughout pregnancy: comparison to salivary cortisol. Physiol Behav. 2011;104(2):348-53.

D.C. Nieman Upper respiratory tract infections and exercise. *Thorax, 50* (1995), pp. 1229-1231

D. Pereg, R. Gow, M. Mosseri, M. Lishner, M. Rieder, S. Van Uum, G. Koren Hair cortisol and the risk for acute myocardial infarction in adult men Stress, 14 (2011), pp. 73-81

Ford, J. L., Boch, S. J., McCarthy, D. O. Feasibility of hair collection for cortisol measurement in population research on adolescent health. *Nurs Res*. 65, (3), 249-255 (2016).

Gerber, M., Brand, S., Lindwall, M., Elliot, C., Kalak, N., & Herrmann, C. (2012). Concerns regarding hair cortisol as a biomarker of chronic stress in exercise and sports science. *Journal of Sports Science and Medicine,11*.

Gleeson, M. (2007). Immune function in sport and exercise. *Journal of Applied Physiology,103*(2), 693-699. doi:10.1152/japplphysiol.00008.2007

Gleeson, M. (2006). Immune system adaptation in elite athletes. Current Opinion in Clinical Nutrition and Metabolic Care, 9(6), 659-665. doi:10.1097/01.mco.0000247476.02650.18

Hill, E. E., Zack, E., Battaglini, C., Viru, M., Viru, A., & Hackney, A. C. (2008). Exercise and circulating Cortisol levels: The intensity threshold effect. *Journal of Endocrinological Investigation, 31*(7), 587-591. doi:10.1007/bf03345606

How much physical activity do children need? (2015, June 04). Retrieved September 22, 2017, from https://www.cdc.gov/physicalactivity/basics/children/index.htm

Lees, C., & Hopkins, J. (2013). Effect of Aerobic Exercise on Cognition, Academic Achievement, and Psychosocial Function in Children: A Systematic Review of Randomized Control Trials. *Preventing Chronic Disease, 10*. doi:10.5888/pcd10.130010

Martin, S. A., Pence, B. D., & Woods, J. A. (2009). Exercise and Respiratory Tract Viral Infections. Exercise Sport Science Rev., 157-164. doi:10.1097/JES.0b013e3181b7b57b

Meyer J, Novak M, Hamel A, Rosenberg K. Extraction and analysis of cortisol from human and monkey hair. J Vis Exp. 2014(83):e50882.

Mikkelsen, K., Stojanovska, L., Polenakovic, M., Bosevski, M., & Apostolopoulus, V. (2017). Exercise and mental health. *Maturitas, 106*, 48-56. doi:https://doi.org/10.1016/j.maturitas.2017.09.003

Norris, R., Carroll, D., & Cochrane, R. (1992). The effects of physical activity and exercise training on psychological stress and well-being in an adolescent population. *Journal of Psychosomatic Research,36*(1), 55-65. doi:10.1016/0022-3999(92)90114-h

Pascoe, M.C., Thompson, D.R., & Ski, C.F. (2017). Yoga, mindfulness-based stress reduction and stress-related physiological measures: A meta-analysis. *Psychoneuroendocrinology, 86*, 152-168. doi: 10.1016/j.psyneuen.2017.08.008.

Petibois, C., Cazorla, G., Poortmans, J., & Dris, G. (2003). Biochemical aspects of overtraining in endurance sports. *Sports Medicine, 33*(2), 83-94. doi:10.2165/00007256-200333020-00001

Salmon, P. (2001). Effects of physical exercise on anxiety, depression, and sensitivity to stress: A unifying theory. *Clinical Psychology Review, 21*(1), 33-61. doi:https://doi.org/10.1016/S0272-7358(99)00032-X

Skoluda N., Dettenborn L., Stalder T., Kirschbaum C. (2012). Elevated hair cortisol concentrations in endurance athletes. Psychoneuroendocrinology 37, 611–617

US Department of Health and Human Services. Physical Activity Guidelines for Americans, 2nd edition. Washington, DC: US Department of Health and Human Services; 2018.

Wester, V. L., & Rossum, E. F. (2015). Clinical applications of cortisol measurements in hair. *European Journal of Endocrinology, 173*(4). doi:10.1530/eje-15-0313

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