



Short Communication

Consistency where it counts: Sleep regularity is associated with circulating white blood cell count in young adults



Elissa K. Hoopes^a, Michele N. D'Agata^a, Felicia R. Berube^a, Sushant M. Ranadive^b,
Freda Patterson^c, William B. Farquhar^a, David G. Edwards^a, Melissa A. Witman^{a,*}

^a Department of Kinesiology and Applied Physiology, College of Health Sciences, University of Delaware, Newark, DE, USA

^b Department of Kinesiology, School of Public Health, University of Maryland, College Park, MD, USA

^c Department of Behavioral Health and Nutrition, College of Health Sciences, University of Delaware, Newark, DE, USA

ARTICLE INFO

Keywords:

Sleep health
Actigraphy
White blood cells
Young adults

ABSTRACT

Background: Sleep irregularity is predictive of poor health outcomes, and particularly those of cardiometabolic origins. The immune system is implicated in the pathogenesis of cardiometabolic diseases, however the relation between sleep regularity and immune cell profile is unclear.

Methods and results: Forty-two healthy young adults (20 ± 2 years) completed 14 days of 24-h wrist actigraphy followed by a morning blood sample to evaluate circulating white blood cells (WBC) and subtypes (neutrophils, lymphocytes, monocytes). Sleep regularity was operationalized as the standard deviation (SD) of nightly sleep duration and SD of sleep onset time. Every 60-min increase in sleep duration SD was associated with an estimated $2.7 \pm 0.60 \times 10^3$ cells/ μL ($p < 0.001$) increase in total WBC count, while every 60-min increase in sleep onset SD was associated with an estimated $2.4 \pm 0.60 \times 10^3$ cells/ μL ($p < 0.001$) increase in WBCs. Sleep duration SD was also associated with lymphocyte count (11.5 ± 3.8 cells/ μL per 1-min increase, $p < 0.01$), while sleep onset SD was associated with neutrophil (34.7 ± 9.8 cells/ μL per 1-min increase, $p < 0.01$) and monocyte counts (3.0 ± 0.9 cells/ μL per 1-min increase, $p < 0.01$). Sleep regularity metrics remained significantly associated with WBCs in a series of regressions which adjusted for sex, body mass index, resting blood pressure, mean sleep duration, physical activity, dietary sodium, and alcohol consumption.

Conclusions: Unfavorable associations between irregular sleep patterns and circulating immune cells are apparent in young adulthood. These findings contribute to the growing body of evidence suggesting that consistent sleep schedules are an important dimension of sleep and circadian health and may reduce excess chronic disease risk.

1. Introduction

The circadian system functions to synchronize physiology, behavior, and anticipated environmental changes across the 24h day (Weaver and Gumz, 2016). Critically, mounting evidence indicates that repeated shifting of the endogenous circadian clock is detrimental to overall health. The most prominent example of this is night-shift work, which is now considered a probable carcinogen (Carcinogenicity of night, 2019) and an independent risk factor for several cardiometabolic conditions including obesity (Sun et al., 2018), hypertension (Manohar et al., 2017), and vascular events (Vyas et al., 2012). Indeed, even mild variations in nightly sleep-wake patterns that are ubiquitous in the general public (Knutson et al., 2007), are linked to circadian disruption, cumulative sleep loss, and increased chronic disease risk (Huang et al., 2020; Huang and Redline, 2019).

The immune system is implicated in the development and progression of several leading causes of death including atherosclerosis, hypertension, and type 2 diabetes (Hansson and Hermansson, 2011; Norlander et al., 2018; Nikolajczyk et al., 2011). One of the most routinely measured and clinically established biomarkers of systemic inflammation and the immune response is circulating white blood cell (WBC) count (Madjid et al., 2004). Higher circulation of total WBCs and subtypes (e.g., neutrophils, lymphocytes, monocytes) are independent predictors of major health outcomes such as hypertension (Shankar et al., 2004), diabetes (Gkrania-Klotsas et al., 2010; Twig et al., 2013), cardiovascular events (Twig et al., 2012), and mortality (Brown et al., 2001; Kabat et al., 2017), even in young adults and those who are disease-free at baseline.

The immune system may represent one physiological pathway linking irregular sleep patterns with negative health outcomes. Experimental

* Corresponding author. STAR Health Sciences Complex, 540 S. College Avenue, Newark, DE, 19716, USA.

E-mail address: mwitman@udel.edu (M.A. Witman).

<https://doi.org/10.1016/j.bbih.2021.100233>

Received 19 December 2020; Received in revised form 20 January 2021; Accepted 24 February 2021

Available online 28 February 2021

2666-3546/© 2021 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

circadian disruption desynchronizes immune functions and promotes inflammation in healthy young volunteers (Cuesta et al., 2016; Morris et al., 2016). Similarly, sleep loss increases peripheral WBC count, with residual elevations even after recovery sleep is obtained (Lasselin et al., 2015; Faraut et al., 2011). To our knowledge, no studies have extended these findings to examine associations between objectively assessed sleep irregularity and circulating WBC count in free-living (i.e., non-laboratory) settings. Additionally, young adults may be susceptible to an adverse immune cell profile, as this age represents a period marked by erratic sleep schedules and frequent sleep loss (Lund et al., 2010).

Thus, we aimed to examine associations between actigraphy-assessed sleep regularity and circulating WBC count in a sample of apparently healthy young adults. We hypothesized that greater variability in sleep duration and sleep onset timing would be independently associated with a greater WBC count in this group.

2. Methods

2.1. Participants

The University of Delaware IRB approved this study. Participants were full-time undergraduate college students (ages 18–25). Individuals were excluded for a history of any major health conditions, sleep disorders, night-shift work, sleep medication use, depression diagnosis or medication use for depression, resting blood pressure (BP) >140/90 mmHg, body mass index (BMI) >34.9 kg/m², current pregnancy, medication use that significantly impacts cardiovascular physiology, or current smoking.

2.2. Sleep assessments

Sleep was assessed with MicroMotionlogger wrist accelerometers worn on the non-dominant wrist for 24-h/day, except during water-based activities, for 14 consecutive days. A conservative threshold of ≥ 12 nights of sleep accelerometry data was required for inclusion in final analyses (Aili et al., 2017). Data were scored with ActionW-2 software (Ambulatory Monitoring, Inc.) using the University of California, San Diego scoring algorithm (Jean-Louis et al., 2001). Participants also completed a standardized nightly sleep diary (Carney et al., 2012).

Sleep duration, or the total time spent asleep from sleep onset to wake onset, was quantified for each night that the accelerometer was worn, and mean sleep duration was generated. Clock time at sleep onset was also quantified for each night, with a mean sleep onset generated for each participant. Sleep duration regularity was represented as the SD of mean sleep duration, while sleep onset regularity was operationalized as the SD in mean sleep onset time across the sleep monitoring period.

2.3. Blood analyses

Following sleep monitoring, participants returned to the lab between 8:00–11:00a.m. for intravenous blood sampling. Participants were fasted and without caffeine, alcohol, heavy exercise for ≥ 12 h, and anti-inflammatory drugs for ≥ 24 h, prior to this visit. Participants were screened for subjective reporting of acute illness prior to blood sampling. Female participants provided samples during the early follicular phase of the menstrual cycle. All data was collected when participants were enrolled in a full-time workload and excluded holidays or final examination periods.

Whole blood was obtained from the antecubital vein after 10-min of supine rest. Total and differential WBC count (including neutrophils, lymphocytes, and monocytes) were determined as part of a clinical complete blood count using automated cell cytometry (Quest Diagnostics, Inc.). 3 mL of whole blood was collected in EDTA-treated vacuum tubes and kept at room temperature until analysis within 24h of sampling. An additional 4 mL of blood was collected in serum separator tubes and used for analysis of fasting lipids and glucose via spectrophotometry.

2.4. Additional measures

BMI (kg/m²) and resting brachial BP were determined (Muntner et al., 2019). Moderate-vigorous physical activity (MVPA) was assessed via torso accelerometry (ActiGraph wgt3x-bt) for 7 consecutive days overlapping with sleep monitoring. Habitual alcohol consumption was quantified via questionnaire and sodium consumption was evaluated using a 3-day diet log, which also overlapped with sleep monitoring.

2.5. Statistical analyses

Bivariate associations between sleep regularity metrics and WBCs were initially examined using unadjusted linear regressions. Additionally, a series of multivariable regression models were tested separately to evaluate the independent associations between sleep regularity metrics with WBCs. Specifically, associations were examined after adjustment for mean sleep duration (Model 1), biological confounders (sex, BMI, resting mean arterial pressure [MAP]; Model 2), and behavioral confounders including MVPA (min/day), dietary sodium consumption (g/day), and alcohol consumption (drinks/week) (Model 3). Regression results are presented as unstandardized coefficients (B) and standard error (SE). Significance was set at $\alpha < 0.05$ (SPSS v. 26.0).

A post hoc power analysis was conducted using G*Power statistical software and revealed that at $\alpha < 0.05$, $n = 42$, and four predictors, statistical power for this study was 0.69 and 0.96 for detecting medium ($f^2 = .15$) and large ($f^2 = .35$) effects, respectively. Thus, there was more than adequate power to detect a large effect, but less than adequate power to detect a small to moderate effect.

3. Results

3.1. Subject characteristics

Forty-two participants (18 males, 24 females) completed sleep monitoring and subsequent blood sampling. By design, participants were generally healthy and exhibited immune cell concentrations within normal ranges. Mean sleep duration was 7.1 ± 0.7 h, while mean sleep onset was $1:20$ a.m. $\pm 1:09$. Mean sleep duration SD was 74 ± 23 min and ranged from 34 to 123, while mean sleep onset SD was 64 ± 21 min and ranged from 30 to 150. Participant characteristics are displayed in Table 1.

3.2. Associations between sleep duration SD and WBC counts

Sleep duration SD was positively associated with total WBC count ($0.05 \pm 0.01 \times 10^3$ cells/ μ L, $p < 0.001$), such that every 60-min increase in sleep duration SD was associated with an estimated $2.7 \pm 0.60 \times 10^3$ cells/ μ L increase in total WBCs. Sleep duration SD was also associated with circulating neutrophils (30.21 ± 11.71 cells/ μ L, $p < 0.01$), lymphocytes (11.46 ± 3.83 cells/ μ L, $p < 0.01$), and monocytes (2.35 ± 1.03 cells/ μ L, $p = 0.01$), all of which remained significant after adjusting for mean sleep duration. Further, after adjusting for sex, BMI, and MAP, the positive associations with total WBC count and lymphocytes remained. Finally, after adjusting for MVPA, alcohol consumption, and sodium consumption, all positive associations between sleep duration SD and WBC counts remained, except sleep duration SD with monocyte count. Multivariable regression results for sleep duration SD are displayed in Table 2.

3.3. Associations between sleep onset SD and WBC counts

Sleep onset SD was positively associated with total WBC count ($0.04 \pm 0.01 \times 10^3$ cells/ μ L, $p < 0.001$), such that every 60-min increase in sleep onset SD was associated with an estimated $2.4 \pm 0.60 \times 10^3$ cells/ μ L increase in total WBCs. Sleep onset SD was also associated with neutrophils (34.68 ± 9.82 cells/ μ L, $p < 0.01$) and monocytes (2.95 ± 0.85 cells/ μ L,

Table 1
Participant characteristics.

Subject Characteristics	Mean±SD
Age, y	21 ± 2
Sex, M/F	18/24
BMI, kg/m ²	23.9 ± 2.5
Systolic BP, mmHg	117 ± 9
Diastolic BP, mmHg	69 ± 7
Heart Rate, bpm	61 ± 10
Blood Chemistry	
Total Cholesterol, mg/dL	160 ± 28
LDL, mg/dL	86 ± 23
HDL, mg/dL	58 ± 13
Triglycerides, mg/dL	74 ± 32
Fasting Glucose, mg/dL	85 ± 6
Total WBC Count, x10 ³ cells/μL (Reference range: 4.50–11.00)	6.10 ± 1.9
Neutrophils, cells/μL (Reference range: 2500–8000)	3385 ± 1819
Lymphocytes, cells/μL (Reference range: 1000–4000)	2038 ± 610
Monocytes, cells/μL (Reference range: 100–700)	495 ± 157
Sleep Metrics	
Sleep Duration, h	7.1 ± 0.7
Sleep Onset, clock time	1:20 a.m. ± 1:09
Sleep Duration SD, min	74 ± 23
Sleep Onset SD, min	64 ± 21
Health Behaviors	
MVPA, min/day	68 ± 24
Dietary Sodium, g/day	3.28 ± 1.40
Alcohol Consumption, drinks/week	6 ± 6

LDL, low-density lipoprotein cholesterol; HDL, high-density lipoprotein.

Table 2
Multivariable regression models of total WBCs and subtypes.

	Model 1	Model 2	Model 3
	B±SE	B±SE	B±SE
Sleep Duration SD, min			
Total WBCs, x10 ³ cells/μL	0.04 ± 0.01**	0.02 ± 0.01*	0.05 ± 0.01**
Neutrophils, cells/μL	29.58 ± 11.90*	11.64 ± 12.45	35.96 ± 12.69**
Lymphocytes, cells/μL	11.01 ± 3.85**	9.92 ± 4.53*	11.73 ± 4.05**
Monocytes, cells/μL	2.17 ± 1.02*	1.31 ± 1.13	1.98 ± 1.07
Sleep Onset SD, min			
Total WBCs, x10 ³ cells/μL	0.04 ± 0.01**	0.03 ± 0.01**	0.04 ± 0.01**
Neutrophils, cells/μL	34.23 ± 10.01**	28.12 ± 9.35**	33.74 ± 11.23**
Lymphocytes, cells/μL	0.84 ± 3.77	0.07 ± 3.99	3.03 ± 4.00
Monocytes, cells/μL	2.79 ± 0.85**	2.17 ± 0.88*	3.01 ± 0.87**

*p<0.05, **p<0.01. Model 1 adjusts for mean sleep duration; Model 2 adjusts for sex, BMI, resting MAP; Model 3 adjusts for MVPA, dietary sodium, and alcohol consumption.

p<0.01) prior to adjustments. After adjusting for mean sleep duration, significant associations between sleep onset SD and total WBCs, neutrophils, and monocytes remained. These associations also persisted after adjusting for sex, BMI, and MAP, and again in models adjusted for MVPA, alcohol consumption, and dietary sodium. Multivariable regression results for sleep onset SD are also displayed in Table 2.

4. Discussion

Irregularity in sleep duration and sleep onset timing were associated with higher circulating WBC count in this sample of healthy young adults. Notably, these associations remained significant after adjusting for confounders, suggestive of an independent link between free-living sleep regularity and circulating immune cells that is apparent even at pre-clinical levels. This presents an intriguing hypothesis that irregular sleep-wake patterns might chronically disrupt the immune system, which could promote disease development and progression if sustained over time. The directionality and physiological underpinnings of these

associations, as well as their impact on disease risk later in life, warrants further investigation in future studies.

Circulating WBC count is predictive of adverse health outcomes including coronary artery disease (Brown et al., 2001), cancers (Shankar et al., 2006), and type 2 diabetes (Gkrania-Klotsas et al., 2010), even in healthy adults demonstrating WBC counts within normal ranges (Twigg et al., 2012, 2013; Brown et al., 2001). In the current study, we observed that every 1-h increase in sleep duration SD was, on average, associated with a ~2700 cells/μL increase in total WBCs, while every 1-h increase in sleep onset SD was associated with an estimated ~2400 cells/μL increase in total WBCs. These associations are particularly notable when considered in the context of prior findings that a total WBC increment of 1000 cells/μL was associated with a 17.4% increase in incident coronary artery disease and a 7.6% increase in incident type 2 diabetes later in life in more than 20,000 healthy young men (Twigg et al., 2012, 2013).

While sleep onset SD and sleep duration SD are both field-based proxies for circadian disruption (Zuraikat et al., 2020), they may also have distinct implications for sleep and circadian health. For example, sleep onset timing is significantly correlated with circadian phase (Martin and Eastman, 2002), therefore a higher sleep onset SD may be illustrating those experiencing greater shifts in endogenous circadian timing. Meanwhile, sleep duration SD represents the magnitude of fluctuations in nightly sleep duration; thus, in addition to shifts in endogenous circadian timing, higher values may also encompass some extent of sleep debt (i.e., those experiencing shortened sleep and ‘catch-up’ sleep episodes). These distinguishing characteristics may have influenced the diverging associations between sleep regularity metrics with WBC subtypes in the current study. Interestingly, while elevations in circulating WBCs are evident following experimental sleep manipulations in healthy adults (Lasselin et al., 2015; Faraut et al., 2011; Born et al., 1997; Boudjeltia et al., 2008), increases in subtypes appear non-specific. For example, some studies inducing sleep loss (e.g., one night of sleep deprivation, several nights of sleep restriction) report increased circulating lymphocytes (Lasselin et al., 2015; Born et al., 1997), while others report no change (Boudjeltia et al., 2008) or even decreases (Faraut et al., 2011). Conversely, elevations in neutrophils and monocytes are more consistently reported (Lasselin et al., 2015; Faraut et al., 2011; Born et al., 1997; Boudjeltia et al., 2008), with sustained elevations in neutrophil count even after recovery sleep is obtained (Lasselin et al., 2015; Faraut et al., 2011). Interpretation of laboratory-based studies are further complicated by varying protocols between studies (i.e., method and duration of sleep manipulation, sex differences between samples, differing times of blood sampling). Additionally, while many experimental protocols inevitably induce variations in sleep duration and/or sleep onset timing over several consecutive nights, they are highly regimented and poorly translate to the varying nature of sleep schedules in free-living settings (Knutson et al., 2007; McMahon et al., 2018). Taken together, our findings highlight the need for future studies to systematically evaluate the distinct sleep and circadian consequences of varying sleep duration versus varying sleep onset as they are experienced in free-living settings, in order to better understand their independent roles in circulating immune cell regulation.

Recent findings from the Multi-Ethnic Study of Atherosclerosis indicated that older adults with the most irregular sleep duration or sleep onset timing demonstrate a >2-fold increased risk of developing CVD over a median follow-up of 4.9 years when compared to those with the most regular sleep patterns (Huang et al., 2020). These sleep regularity metrics were also shown to independently predict the clustering of multiple metabolic abnormalities in a sample from the same cohort (Huang and Redline, 2019). The aforementioned associations were extensively adjusted for traditional CVD risk factors, demographics, and other sleep characteristics, yet these covariates were only able to explain a marginal portion of the associations between sleep regularity and cardiometabolic risk. However, neither study adjusted for immune biomarkers, highlighting the possibility that the immune system may have mediated a portion of that risk.

Identifying a link between free-living sleep regularity and the immune system in young adulthood has considerable implications for population health, as it builds upon literature that has largely focused on older adults. Indeed, irregular sleep-wake patterns have been independently linked to cardiometabolic morbidities and risk factors including metabolic syndrome (Huang and Redline, 2019), hypertension (Brindle et al., 2019), adiposity (Ogilvie et al., 2016), and insulin resistance (Taylor et al., 2016) in midlife or later. Albeit less frequently examined, youth and young adults with irregular sleep patterns also tend to have poorer cardiometabolic health, such as increased adiposity (Nicholson et al., 2020; He et al., 2015), impaired microvascular function, (Hoopes et al., 2021) and poorer diet (Ogilvie et al., 2018) when compared to those with regular sleep patterns. Our findings also parallel two studies reporting independent associations between sleep duration SD and elevated C-reactive protein in children (Nielsen et al., 2016) and adolescents (Park et al., 2016). Considering sleep habits in adults tend to persist over time (Knutson et al., 2007; McMahon et al., 2018), establishing consistent sleep-wake schedules early in life represents an upstream intervention target that could assist in long-term prevention of inflammatory-mediated chronic conditions, and particularly cardiometabolic diseases.

The lack of determinable directionality, the inability to examine sex-specific associations, and our homogenous population are study limitations. Additionally, the small sample size limited our ability to include all covariates of interest into one model. However, it is important to note that associations across regression models were mostly consistent and are in agreement with previous literature on sleep irregularity and cardiometabolic health in larger and more diverse samples (Huang et al., 2020; Park et al., 2016; Brindle et al., 2019), supporting the validity of our results. Considering blood samples were only taken at one timepoint, future studies should extend these findings by evaluating diurnal rhythms in circulating WBCs in the context of sleep irregularity. Evaluation of WBC functions (i.e., spontaneous activation patterns, response to stimulation) are also needed to identify immune cell phenotypes associated with sleep irregularity. Study strengths include consideration of several important confounds and a robust actigraphy protocol with minimal missing datapoints.

In summary, greater variations in nightly sleep duration and sleep onset time are significantly associated with higher WBC counts in apparently healthy young adults. This suggests that the suboptimal immune responses to experimental circadian misalignment and sleep loss with recovery may also be apparent with fluctuating sleep schedules in free-living settings. Our results imply that if these sleep patterns become chronic, there may be an increased risk for inflammatory-mediated health conditions with advancing age. Longitudinal studies are needed to systematically examine this hypothesis. Additionally, interventions are needed to evaluate the health benefits of consistent sleep patterns initiated in young adulthood.

Sources of funding

This study was supported, in part, by NIH-NIGMS P20GM113125 (MAW).

Declaration of competing interest

There is no relationship with industry or conflict of interest.

Acknowledgements

The authors thank Wendy Nichols, RN, and the Nurse Managed Primary Care Center for their assistance with blood collection and processing.

References

- Aili, K., Astrom-Paulsson, S., Stoetzer, U., Svartengren, M., Hillert, L., 2017. Reliability of actigraphy and subjective sleep measurements in adults: the design of sleep assessments. *J. Clin. Sleep Med.* 13, 39–47.
- Born, J., Lange, T., Hansen, K., Mölle, M., Fehm, H.L., 1997. Effects of sleep and circadian rhythm on human circulating immune cells. *J. Immunol.* 158, 4454–4464.
- Boudjeltia, K.Z., Faraut, B., Stenuit, P., Esposito, M.J., Dyzma, M., Brohee, D., Ducobu, J., Vanhaeverbeek, M., Kerkhofs, M., 2008. Sleep restriction increases white blood cells, mainly neutrophil count, in young healthy men: a pilot study. *Vasc. Health Risk Manag.* 4, 1467–1470.
- Brindle, R.C., Yu, L., Buysse, D.J., Hall, M.H., 2019. Empirical derivation of cutoff values for the sleep health metric and its relationship to cardiometabolic morbidity: results from the midlife in the United States (midus) study. *Sleep*.
- Brown, D.W., Giles, W.H., Croft, J.B., 2001. White blood cell count: an independent predictor of coronary heart disease mortality among a national cohort. *J. Clin. Epidemiol.* 54, 316–322.
- Carcinogenicity of Night Shift Work, vol. 20, 2019. International Agency for Research on Cancer Working Group, pp. 1058–1059.
- Carney, C.E., Buysse, D.J., Ancoli-Israel, S., Edinger, J.D., Krystal, A.D., Lichstein, K.L., Morin, C.M., 2012. The consensus sleep diary: standardizing prospective sleep self-monitoring. *Sleep* 35, 287–302.
- Cuesta, M., Boudreau, P., Dubreau-Laramée, G., Cermakian, N., Boivin, D.B., 2016. Simulated night shift disrupts circadian rhythms of immune functions in humans. *J. Immunol.* 196, 2466–2475.
- Faraut, B., Boudjeltia, K.Z., Dyzma, M., Rousseau, A., David, E., Stenuit, P., Franck, T., Van Antwerpen, P., Vanhaeverbeek, M., Kerkhofs, M., 2011. Benefits of napping and an extended duration of recovery sleep on alertness and immune cells after acute sleep restriction. *Brain Behav. Immun.* 25, 16–24.
- Gkrania-Klotsas, E., Ye, Z., Cooper, A.J., Sharp, S.J., Luben, R., Biggs, M.L., Chen, L.K., Gokulakrishnan, K., Hanefeld, M., Ingelsson, E., Lai, W.A., Lin, S.Y., Lind, L., Lohsoonthorn, V., Mohan, V., Muscarì, A., Nilsson, G., Ohrvik, J., Chao Qiang, J., Jenny, N.S., Tamakoshi, K., Temelkova-Kurktschiev, T., Wang, Y.Y., Yajnik, C.S., Zoli, M., Khaw, K.T., Forouhi, N.G., Wareham, N.J., Langenberg, C., 2010. Differential white blood cell count and type 2 diabetes: systematic review and meta-analysis of cross-sectional and prospective studies. *PLoS One* 5, e13405.
- Hansson, G.K., Hermansson, A., 2011. The immune system in atherosclerosis. *Nat. Immunol.* 12, 204–212.
- He, F., Bixler, E.O., Liao, J., Berg, A., Imamura Kawasawa, Y., Fernandez-Mendoza, J., Vgontzas, A.N., Liao, D., 2015. Habitual sleep variability, mediated by nutrition intake, is associated with abdominal obesity in adolescents. *Sleep Med.* 16, 1489–1494.
- Huang, T., Redline, S., 2019. Cross-sectional and prospective associations of actigraphy-assessed sleep regularity with metabolic abnormalities: the multi-ethnic study of atherosclerosis. *Diabetes Care* 42, 1422–1429.
- Hoopes, E.K., Berube, F.R., D'Agata, M.N., Patterson, F., Farquhar, W.B., Edwards, D.G., Witman, M.A., 2021. Sleep duration regularity, but not sleep duration, is associated with microvascular function in college students. *Sleep* 44 (2), zsa175.
- Huang, T., Mariani, S., Redline, S., 2020. Sleep irregularity and risk of cardiovascular events: the multi-ethnic study of atherosclerosis. *J. Am. Coll. Cardiol.* 75, 991–999.
- Jean-Louis, G., Kripke, D.F., Mason, W.J., Elliott, J.A., Youngstedt, S.D., 2001. Sleep estimation from wrist movement quantified by different actigraphic modalities. *J. Neurosci. Methods* 105, 185–191.
- Kabat, G.C., Kim, M.Y., Manson, J.E., Lessin, L., Lin, J., Wassertheil-Smoller, S., Rohan, T.E., 2017. White blood cell count and total and cause-specific mortality in the women's health initiative. *Am. J. Epidemiol.* 186, 63–72.
- Knutson, K.L., Rathouz, P.J., Yan, L.L., Liu, K., Lauderdale, D.S., 2007. Intra-individual daily and yearly variability in actigraphically recorded sleep measures: the cardia study. *Sleep* 30, 793–796.
- Lasselin, J., Rehman, J.U., Akerstedt, T., Lekander, M., Axelsson, J., 2015. Effect of long-term sleep restriction and subsequent recovery sleep on the diurnal rhythms of white blood cell subpopulations. *Brain Behav. Immun.* 47, 93–99.
- Lund, H.G., Reider, B.D., Whiting, A.B., Prichard, J.R., 2010. Sleep patterns and predictors of disturbed sleep in a large population of college students. *J. Adolesc. Health* 46, 124–132.
- Madjid, M., Awan, I., Willerson, J.T., Casscells, S.W., 2004. Leukocyte count and coronary heart disease: implications for risk assessment. *J. Am. Coll. Cardiol.* 44, 1945–1956.
- Manohar, S., Thongprayoon, C., Cheungpasitporn, W., Mao, M.A., Herrmann, S.M., 2017. Associations of rotational shift work and night shift status with hypertension: a systematic review and meta-analysis. *J. Hypertens.* 35, 1929–1937.
- Martin, S.K., Eastman, C.I., 2002. Sleep logs of young adults with self-selected sleep times predict the dim light melatonin onset. *Chronobiol. Int.* 19, 695–707.
- McMahon, D.M., Burch, J.B., Wirth, M.D., Youngstedt, S.D., Hardin, J.W., Hurley, T.G., Blair, S.N., Hand, G.A., Shook, R.P., Drenowatz, C., Burgess, S., Hebert, J.R., 2018. Persistence of social jetlag and sleep disruption in healthy young adults. *Chronobiol. Int.* 35, 312–328.
- Morris, C.J., Purvis, T.E., Hu, K., Scheer, F.A.J.L., 2016. Circadian misalignment increases cardiovascular disease risk factors in humans. *Proc. Natl. Acad. Sci. U. S. A.* 113, E1402–E1411.
- Muntner, P., Shimbo, D., Carey, R.M., Charleston, J.B., Gaillard, T., Misra, S., Myers, M.G., Ogedegbe, G., Schwartz, J.E., Townsend, R.R., Urbina, E.M., Viera, A.J., White, W.B., Wright Jr., J.T., 2019. Measurement of blood pressure in humans: a scientific statement from the american heart association. *Hypertension* 73, e35–e66.

- Nicholson, L.M., Egbert, A.H., Moreno, J.P., Bohnert, A.M., 2020. Variability of sleep and relations to body weight among first-year college students. *Int. J. Behav. Med.*
- Nielsen, M.S., Quist, J.S., Chaput, J.P., Dalskov, S.M., Damsgaard, C.T., Ritz, C., Astrup, A., Michaelsen, K.F., Sjoedin, A., Hjorth, M.F., 2016. Physical activity, sedentary time, and sleep and the association with inflammatory markers and adiponectin in 8- to 11-year-old Danish children. *J. Phys. Activ. Health* 13, 733–739.
- Nikolajczyk, B.S., Jagannathan-Bogdan, M., Shin, H., Gyurko, R., 2011. State of the union between metabolism and the immune system in type 2 diabetes. *Gene Immun.* 12, 239–250.
- Norlander, A.E., Madhur, M.S., Harrison, D.G., 2018. The immunology of hypertension. *J. Exp. Med.* 215, 21–33.
- Ogilvie, R.P., Redline, S., Bertoni, A.G., Chen, X., Ouyang, P., Szklo, M., Lutsey, P.L., 2016. Actigraphy measured sleep indices and adiposity: the multi-ethnic study of atherosclerosis (mesa). *Sleep* 39, 1701–1708.
- Ogilvie, R.P., Lutsey, P.L., Widome, R., Laska, M.N., Larson, N., Neumark-Sztainer, D., 2018. Sleep indices and eating behaviours in young adults: findings from project eat. *Publ. Health Nutr.* 21, 689–701.
- Park, H., Tsai, K.M., Dahl, R.E., Irwin, M.R., McCreath, H., Seeman, T.E., Fuligni, A.J., 2016. Sleep and inflammation during adolescence. *Psychosom. Med.* 78, 677–685.
- Shankar, A., Klein, B.E., Klein, R., 2004. Relationship between white blood cell count and incident hypertension. *Am. J. Hypertens.* 17, 233–239.
- Shankar, A., Wang, J.J., Rohtchina, E., Yu, M.C., Kefferd, R., Mitchell, P., 2006. Association between circulating white blood cell count and cancer mortality: a population-based cohort study. *Arch. Intern. Med.* 166, 188–194.
- Sun, M., Feng, W., Wang, F., Li, P., Li, Z., Li, M., Tse, G., Vlaanderen, J., Vermeulen, R., Tse, L.A., 2018. Meta-analysis on shift work and risks of specific obesity types. *Obes. Rev.* 19, 28–40.
- Taylor, B.J., Matthews, K.A., Hasler, B.P., Roeklein, K.A., Kline, C.E., Buysse, D.J., Kravitz, H.M., Tian, A.G., Harlow, S.D., Hall, M.H., 2016. Bedtime variability and metabolic health in midlife women: the swan sleep study. *Sleep* 39, 457–465.
- Twig, G., Afek, A., Shams, A., Derazne, E., Tzur, D., Gordon, B., Tirosh, A., 2012. White blood cell count and the risk for coronary artery disease in young adults. *PLoS One* 7, e47183.
- Twig, G., Afek, A., Shams, A., Derazne, E., Tzur, D., Gordon, B., Tirosh, A., 2013. White blood cells count and incidence of type 2 diabetes in young men. *Diabetes Care* 36, 276–282.
- Vyas, M.V., Garg, A.X., Iansavichus, A.V., Costella, J., Donner, A., Laugsand, L.E., Janszky, I., Mrkobrada, M., Parraga, G., Hackam, D.G., 2012. Shift work and vascular events: systematic review and meta-analysis. *BMJ* 345, e4800.
- Weaver, D.R., 2016. Introduction to circadian rhythms and mechanisms of circadian oscillations. In: Gumz, M.L. (Ed.), *Circadian Clocks: Role in Health and Disease*. Springer, New York, NY.
- Zuraikat, F.M., Makarem, N., Redline, S., Aggarwal, B., Jelic, S., St-Onge, M.P., 2020. Sleep regularity and cardiometabolic health: is variability in sleep patterns a risk factor for excess adiposity and glycemic dysregulation? *Curr. Diabetes Rep.* 20, 38.