# UNIVERSITY OF SÃO PAULO SCHOOL OF ARTS, SCIENCES AND HUMANITIES GRADUATE PROGRAM IN COMPLEX SYSTEMS MODELING

Daniel Kachvartanian de Azevedo

Is latitude associated with chronotype?

São Paulo

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# **Original version**

Thesis presented to the School of Arts, Sciences and Humanities at the University of São Paulo, as a requirement for the degree of Master of Science by the Graduate Program in Complex Systems Modeling.

Area of concentration: Complex Systems.

Supervisor: Prof. Dr. Camilo Rodrigues Neto

São Paulo

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#### **ABSTRACT**

Vartanian, D. (2024). *Is latitude associated with chronotype?* [Master's Thesis, University of São Paulo].

Theories on circadian rhythms are well-established in science, but there is still a need to test them in larger samples to gain a better understanding of the expression of temporal phenotypes. This thesis investigates the hypothesis that latitude influences chronotype expression, based on the idea that regions closer to the poles receive less sunlight over the year than equatorial regions. This difference suggests that equatorial areas have a stronger solar zeitgeber, which could lead to greater synchronization of circadian rhythms with the light-dark cycle, reducing the amplitude and diversity of circadian phenotypes, resulting in a higher propensity for morningness in those populations. To test this hypothesis, data from 120, 265 individuals from all regions of Brazil were analyzed, collected in 2017 based on the Munich ChronoType Questionnaire (MCTQ), a validated questionnaire to assess circadian phenotypes derived from individuals' sleepwake cycles over the past four weeks. The analysis, using nested linear regression models, revealed a negligible effect of latitude on the variation in chronotype expression (Cohen's  $f^2$ : 0.00740), contrasting with recent studies. Although the hypothesis is reasonable and aligns with evolutionary theories of temporal biological systems, the results suggest that the phenomenon of entrainment is more complex than previously thought.

**Keywords**: Complexity science. Complex systems. Chronobiology. Biological rhythms. Chronotypes. Circadian phenotypes. Sleep. Entrainment. Latitude. MCTQ.

#### **RESUMO**

Vartanian, D. (2024). A latitude está associada ao cronotipo? [Dissertação de Mestrado, Universidade de São Paulo].

As teorias sobre ritmos circadianos estão bem estabelecidas na ciência, mas ainda há a necessidade de testá-las em amostras mais amplas para compreender melhor a expressão dos fenótipos temporais. Esta dissertação investiga a hipótese de que a latitude influencia a expressão dos cronotipos, baseada na ideia de que regiões próximas aos polos recebem menos luz solar ao longo do ano do que as regiões equatoriais. Esse diferencial sugere que áreas equatoriais possuem um zeitgeber solar mais forte, o que poderia levar a uma maior sincronização dos ritmos circadianos com o ciclo claroescuro, reduzindo a amplitude e a diversidade de fenótipos circadianos, resultando em uma propensão maior ao cronotipo matutino. Para testar essa hipótese, foram analisados dados de 120.265 indivíduos de todas as regiões do Brasil, coletados em 2017 com base no Munich ChronoType Questionnaire (MCTQ), um questionário validado para medir fenótipos circadianos a partir do ciclo sono-vigília nas últimas quatro semanas. A análise, utilizando modelos de regressão linear aninhados, revelou um efeito negligenciável da latitude na variação da expressão dos cronotipos ( $f^2$  de Cohen: 0,00740), em contraste com estudos recentes. Embora a hipótese faça sentido e esteja alinhada com teorias evolutivas dos sistemas biológicos temporais, os resultados sugerem que o fenômeno de entraiment é mais complexo do que se imaginava.

**Palavras-chaves**: Ciência da complexidade. Sistemas complexos. Cronobiologia. Ritmos biológicos. Cronotipos. Fenótipos circadianos. Sono. Entrainment. Latitude. MCTQ.

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#### 1 INTRODUCTION

# Important

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This chapter is currently a dumping ground for ideas, and I don't recommend reading it.

You are currently viewing the preliminary print version of this master's thesis.

This document follows the collection of articles thesis format. This first chapter serves as an introduction to the thesis subject, providing also its justification and aims. The subsequent chapters consist of a series of essays and a article connected to the thesis, with the exception of the last one, which encompasses a general discussion and final remarks.

All analyses in this document are reproducible and were conducted using the R programming language along with the Quarto publishing system. It's worth noting that this type of thesis is best suited for online viewing. To access the digital version and see the latest research updates, please visit https://danielvartan.github.io/mastersthesis/.

All of these projects are developed using secure, open-source tools and adhere to the best international standards. They are designed to ensure 100% reproducibility and are accompanied by extensive documentation.

# 1.1 AIMS

This thesis focuses on the expression of sleep and circadian phenotypes (chronotypes) with the aim of providing answers to the following questions:

- 1. How are the sleep-wake cycles and circadian phenotypes of the adult Brazilian population characterized?
- 2. Is latitude associated with the regulation of circadian rhythms in humans?

The basic hypothesis to be tested is that populations residing near the equator (latitude 0°) have, on average, a shorter/more morning-oriented circadian phenotype compared to populations living near the Earth's poles (H1) (Hut et al., 2013; Leocadio-Miguel et al., 2014, 2017; Pittendrigh et al., 1991; Randler, 2008; Randler & Rahafar, 2017; Roenneberg, Wirz-Justice, & Merrow, 2003).

The primary objectives (PO) are as follows:

- A) Quantitatively describe the expression of sleep-wake cycles and circadian phenotypes of the Brazilian adult population at the end of the year 2017 (pre-pandemic).
- B) Investigate and model the presence/absence of a significant association and effect between decimal degrees of latitude (independent variable (IV)) and circadian phenotypes (dependent variable (DV)) of the Brazilian population.

To achieve the primary objectives, the following secondary objectives (SO) have been outlined:

- i) Conduct data cleaning, validation, and transformation processes on the obtained sample data.
- ii) Collect secondary data on geolocation and solarimetric models and crossreference them with the primary data.
- iii) Develop algorithms for generating randomly sampled subsets adjusted to the proportions of the analyzed Brazilian regions, based on the latest Brazilian demographic census.
- iv) Develop algorithms and models to help with the processing of MCTQ data and to simulate the complexity of the entrainment phenomena.
- v) Evaluate and discuss the presence/absence of significant differences in the values of the local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days (MSF<sub>sc</sub>), MCTQ proxy for measuring the chronotype, based on decimal degrees of latitude (IV), while controlling for known covariates such as subjects' gender and age.

# 1.2 THESIS STRUCTURE

# Three essays

The main project is the test of the latitude hypothesis, which serves as the primary investigation. It's important to note that all the preceding projects converge into this one. The first project focuses on validating the MCTQ translation used for data collection. The

second project involves the development of data processing tools. The third project is responsible for the necessary data manipulation to prepare it for analysis. The fourth project aims to offer valuable insights and guidance for the upcoming tasks.

#### 2 ON CHRONOBIOLOGY

# Important

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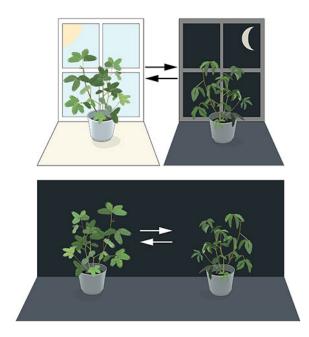
The dimension of time, manifest in the form of rhythms and cycles, like the alternating patterns of day and night as well as the annual transition of seasons, was consistently featured in the evolutionary journey of not only the human species but also all other life forms on our planet. These rhythms and cycles brought with them evolutionary pressures, resulting in the development of a temporal organization that allowed organisms to survive and reproduce in response to the conditions imposed by the environments they inhabited (Menna-Barreto, 2003; Pittendrigh, 1981). An example of this organization can be observed in the presence of different activity-rest patterns among living beings as they adapt to certain temporal niches, such as the diurnal behavior of humans and the nocturnal behavior of cats and some rodents (Foster & Kreitzman, 2005).

For years, scientists debated whether this organization was solely in response to environmental stimuli or if it was also present endogenously, internally, within organisms (Rotenberg et al., 2003). One of the early seminal studies describing a potential endogenous rhythmicity in living beings was conducted in 1729 by the French astronomer Jean Jacques d'Ortous de Mairan. De Mairan observed the movement of the sensitive plant (*mimosa pudica*) by isolating it from the light-dark cycle and found that the plant continued to move its leaves periodically (Figure 1) (de Mairan, 1729; Rotenberg et al., 2003). Charles Darwin also also wrote about the movement observed in plants and made thematic explorations of these intriguing "periodical phenomena"(Andrade & Beale, 2024). The search for this internal timekeeper in living beings only began to solidify in the 20th century through the efforts of scientists like Jürgen Aschoff, Colin Pittendrigh, Franz Halberg, and Erwin Bünning, culminating in the establishment of the science known as chronobiology<sup>1</sup>, with a significant milestone being the Cold Spring Harbor Symposium on Quantitative Biology: Biological Clocks in 1960 (*chrónos*, from

<sup>&</sup>lt;sup>1</sup> Some say the term *chronobiology* was coined by Franz Halberg during the Cold Spring Harbor Symposium on Quantitative Biology, vol. XXV (Menna-Barreto & Marques, 2023, p. 21).

Greek, meaning time; and *biology*, pertaining to the study of life) (Laboratory, n.d.; Rotenberg et al., 2003). However, the recognition of endogenous rhythmicity by the global scientific community truly came in 2017 when Jeffrey Hall, Michael Rosbash, and Michael Young were awarded the Nobel Prize in Physiology or Medicine for their discoveries of molecular mechanisms that regulate the circadian rhythm in fruit flies (*circā*, from Latin, meaning around, and *dĭes*, meaning day (Latinitium, n.d.) – a rhythm that expresses itself in approximately one day) (Nobel Prize Outreach AB, n.d.).

Figure 1 – Illustration of a circadian rhythm in the movement of the leaves of the sensitive plant (mimosa pudica) observed by Jacques d'Ortous de Mairan in 1729.

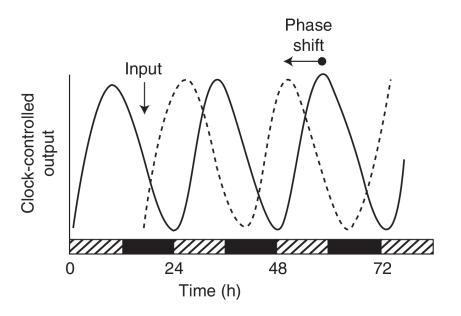


Source: Reproduction from Nobel Prize Outreach AB (n.d.).

Science has already demonstrated and described various biological rhythms and their impacts on organisms. These rhythms can occur at different levels, whether at a macro level, such as the menstrual cycle, or even at a micro level, such as rhythms expressed within cells (Roenneberg & Merrow, 2016). Like many other biological phenomena, these are complex systems present in all living beings, i.e., a emergence created by a large number of connected and interecticve agents that exhibit adaptive characteristics, all without the need of a central control (Boccara, 2010). It is understood today that the endogeneity of rhythms has provided organisms with an anticipatory capacity, allowing them to organize resources and activities before they are needed (Marques & Menna-Barreto, 2003).

Despite the endogenous nature of these rhythms, they can still be regulated by the external environment. Signals (cues) from the environment that occur cyclically and have the ability to regulate biological rhythmic expression are called zeitgebers (from the German *zeit*, meaning time, and *geber*, meaning donor (Cambridge University Press, n.d.)). These zeitgebers act as synchronizers by entraining the phases of biological rhythms (Khalsa et al., 2003; Kuhlman et al., 2018) (see Figure 2). Among the known zeitgebers are, for example, meal timing and changes in environmental temperature (Aschoff, 1981; Roenneberg & Merrow, 2016). However, the most influential of them is the light-dark cycle. It is understood that the day/night cycle, resulting from the rotation of the Earth, has provided the vast majority of organisms with an oscillatory system with a periodic duration of approximately 24 hours (Kuhlman et al., 2018; Roenneberg, Kuehnle, et al., 2007).

Figure 2 – Illustration of a circadian rhythm (output) whose phase is entrained in the presence of a zeitgeber (input). The rectangles represent the light-dark cycle.



Source: Adapted from Kuhlman et al. (2018).

Naturally, the expression of this temporal organization varies from organism to organism, even among members of the same species, whether due to the different ways they are exposed to the environment or the differences in the expression of endogenous rhythmicity, which, in turn, results from gene expression (Roenneberg, Kumar, & Merrow, 2007). The interaction between these two expressions, external and internal,

of the environment and genotype, generates a signature, an observable characteristic, which is called a phenotype (Frommlet et al., 2016).

The various temporal characteristics of an organism can be linked to different oscillatory periods. Among these are circadian phenotypes, which refer to characteristics observed in rhythms with periods lasting about a day (Foster & Kreitzman, 2005). Another term used for these temporal phenotypes, as the name suggest, is *chronotype* (Ehret, 1974; Pittendrigh, 1993). This term is also often used to differentiate phenotypes on a spectrum ranging from morningness to eveningness (Horne & Ostberg, 1976; Roenneberg, Wirz-Justice, et al., 2019).

Sleep is a phenomenon that exhibits circadian expression. By observing the sleep characteristics of individuals, it is possible to assess the distribution of circadian phenotypes within the same population, thereby investigating their covariates and other relevant associations (Roenneberg, Wirz-Justice, & Merrow, 2003). This is because sleep regulation is understood as the result of the interaction between two processes: a homeostatic process (referred to as the S process), which is sleep-dependent and accumulates with sleep deprivation, and a circadian process (referred to as the C process), whose expression can be influenced by zeitgebers, such as the light-dark cycle (Borbély, 1982; Borbély et al., 2016) (Figure 3 illustrates these two process). Considering that the circadian rhythm (the C process) is present in sleep, its characteristics can be estimated if the S process can be controlled.

Awake Sleeping

Awake Sleeping

6 16 23 4 6 16 23 4 6

Time (h)

Figure 3 – Illustration of the interaction of the S process and the C process in sleep regulation. The figure depicts two scenarios: one without sleep deprivation and another with sleep deprivation. The *y*-axis represents the level of the process.

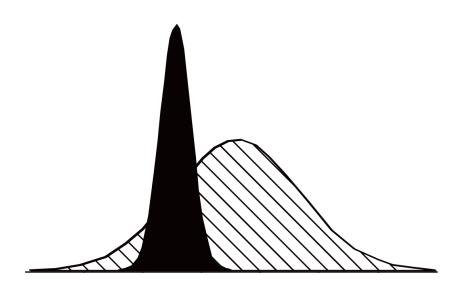
Source: Adapted from Borbély (1982).

Although many theories related to sleep and circadian rhythms are well-established in science, it is still necessary to verify and test them in larger samples to obtain a more accurate picture of the mechanisms related to the ecology of sleep and chronotypes. This project undertakes this commitment with the aim of investigating a hypothesis that is still relatively untested but widely accepted in chronobiology, which suggests that latitude is associated with the regulation of circadian rhythms (Hut et al., 2013; Leocadio-Miguel et al., 2014, 2017; Pittendrigh et al., 1991; Randler, 2008; Randler & Rahafar, 2017; Roenneberg, Wirz-Justice, & Merrow, 2003).

The latitude hypothesis is based on the idea that regions located at latitudes close to the poles, on average, experience less annual sunlight exposure compared to regions near the equator. Therefore, it is deduced that regions near latitude 0° have a stronger solar zeitgeber, which, according to chronobiology theories, should lead to a greater propensity for the synchronization of circadian rhythms in these populations with the light-dark cycle. This would reduce the amplitude and diversity of circadian phenotypes found due to a lower influence of individuals' characteristic endogenous periods (Figure 4 illustrates this effect). This would also give these populations a morningness characteristic when compared to populations living farther from the equator,

where the opposite would occur – greater amplitude and diversity of circadian phenotypes and an eveningness characteristic compared to populations living near latitude 0° (Roenneberg, Wirz-Justice, & Merrow, 2003).

Figure 4 – Different chronotype distributions, influenced by strong and weak zeitgebers – black for strong and hatched for weak. An illustration of the effect hypothesized by the latitude hypothesis.



Source: Adapted from Roenneberg, Wirz-Justice, and Merrow (2003).

To achieve the mentioned objectives, this project will rely on a dataset of the sleep-wake cycle expression of the Brazilian population, consisting of 120,265 subjects covering all states of the country. This dataset was collected in 2017 and is based on the Munich ChronoType Questionnaire (MCTQ), a widely validated scale used to measure chronotypes based on individuals' sleep-wake cycle expression in the last four weeks (Roenneberg, Wirz-Justice, & Merrow, 2003; Roenneberg et al., 2012).

# **3 ON COMPLEXITY SCIENCE**

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# 4 ON THE LATITUDE HYPOTHESIS

# Important

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#### IS LATITUDE ASSOCIATED WITH CHRONOTYPE?

# Note

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This chapter should be readable but is currently undergoing final polishing.

# Warning

The results shown here are **preliminary**, so please take them with a grain of salt.

The data has not yet been fully cleaned, balanced, and cross-referenced with the secondary databases. Think of these results as a low-resolution preview of the final results. The step-by-step analysis can be seen in the appendices section.

# Target journal

1. Scientific Reports (IF 2022: 4.6/JCR | A1/2017-2020).

# Note

The following study was performed by Daniel Vartanian (DV), Mario Pedrazzoli (MP) and Camilo Rodrigues Neto (CR).

**DV** contributed to the design and implementation of the study. **DV** and **MP** collected the data. **DV** and **CR** performed the statistical analysis. **DV** wrote the manuscript. All authors discussed the results and revised the final manuscript.

Future reference: Vartanian, D., Pedrazzoli, M., & Rodrigues Neto, C. (2024). A biological approach for the latitudinal cline of the chronotype. Scientific Reports.

Chronotypes are temporal phenotypes (Ehret, 1974; Pittendrigh, 1993). Observable traits, like weight and eye color. Our current understanding of these traits is that they are linked to our environment and are the result of evolution pressures for creating an inner temporal organization (Aschoff, 1989; Paranipe & Sharma, 2005), a way that organisms found to anticipate events. Having such an important function in nature, these internal rhythms need to be closely aligned with environmental changes. The agents that shift these oscillations towards the environment are called zeitgebers and the shift phenomenon is called entrainment (Roenneberg, Daan, & Merrow, 2003; Roenneberg et al., 2010). The main zeitgeber for humans is light exposure, particularly the light of the sun (Khalsa et al., 2003; Minors et al., 1991; Roenneberg, Kumar, & Merrow, 2007). Considering the major role of light on entrainment, several studies hypothesized that the latitude shift of the sun could influence or even define the chronotypes of different populations (Horzum et al., 2015; Hut et al., 2013; Leocadio-Miguel et al., 2014, 2017; Pittendrigh et al., 1991; Randler & Rahafar, 2017). For example, populations that live close to the equator would be, on average, more entrained to the light-dark cycle and have morning-leaning characteristics. Here we test this hypothesis using a biological measure, the chronotype state, provided by the Munich ChronoType Questionnaire (Roenneberg, Wirz-Justice, & Merrow, 2003). We tested the latitude hypothesis on a sample with 76,744 subjects living in different latitudes in Brazil. Our results show that, even with a wide, big, and aligned sample, the latitude is associated only with negligible effect sizes. The entrainment phenomenon appears to be much more complex than previously imagined, opening new questions and contradictions that need to be further investigated.

# 5.1 MAIN TEXT

#### 5.1.1 **Introduction**

Humans can differ from one another in many ways. These observable traits, like hair color or height, are called phenotypes and are also presented in the way that our body functions.

A chronotype is a temporal phenotype (Ehret, 1974; Pittendrigh, 1993). This word is usually used to refer to endogenous circadian rhythms, i.e., rhythms which periods that are close to a day or 24 hours (*circa diem*). The current body of knowledge of Chronobiology, the science that studies biological rhythms, indicates that the evolution of these internal oscillators is linked to our oscillatory environment, like the day and night cycle, which, along with our evolution, created environmental pressures for the development of a temporal organization (Aschoff, 1989; Paranjpe & Sharma, 2005). A way in which an organism could predict events and better manage its needs, like storing food for the winter.

A temporal system wouldn't be of much use if it could not follow environmental changes. To those environmental signals that can regulate the biological rhythms are

given the name zeitgeber (from the German Zeit, time, and Geber, giver). These zeitgebers produce inputs in our bodies that can shift and align those rhythms. This phenomenon is called entrainment (Roenneberg, Daan, & Merrow, 2003; Roenneberg et al., 2010).

The main zeitgeber known today is the light, particularly the sun's light (Khalsa et al., 2003; Minors et al., 1991; Roenneberg, Kumar, & Merrow, 2007). Considering its influence in entraining the biological temporal system, several studies hypothesize that the latitudinal shift of the sun, related to the earth's axis, would produce, on average, different temporal traits in populations that live close to the equator line when compared to populations that live close to the planet's poles (Horzum et al., 2015; Hut et al., 2013; Leocadio-Miguel et al., 2014, 2017; Pittendrigh et al., 1991; Randler & Rahafar, 2017). That is because the latter ones would have greater oscillations in sun activity and an overall weak solar zeitgeber. This is the latitude hypothesis, that can also appear as the environmental hypothesis of circadian rhythm regulation.

Recently there have been attempts to test the latitude hypothesis in different settings, but, at least in humans, none of them have been successful in seeing a significant effect size related to the latitudinal cline. Some of these approaches worked with secondary data and with small samples. One of the most serious attempts of testing this hypothesis was made by Leocadio-Miguel et al. (2017). They measured the chronotype of 12,884 Brazillian subjects on a wide latitudinal spectrum using the Morningness–Eveningness Questionnaire (MEQ). Their results showed a negligible effect size. One possible reason for this is that the MEQ measures psychological traits and not biological states (Roenneberg, Pilz, et al., 2019), i.e., the circadian oscillation itself, therefore, it's not the best way to answer the question (Leocadio-Miguel et al., 2014).

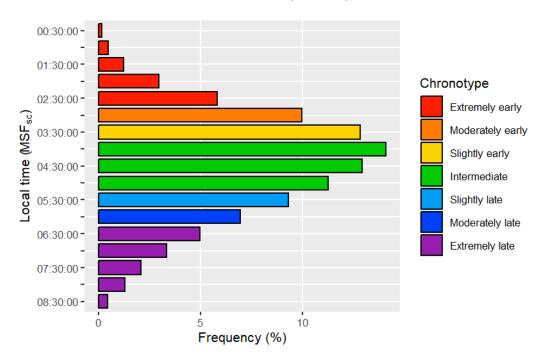
This article brings a novel attempt to test the latitude hypothesis, using, this time, a biological approach provided by the Munich ChronoType Questionnaire (MCTQ) (Roenneberg, Wirz-Justice, & Merrow, 2003). Furthermore, the test was carried out on the biggest chronotype sample ever collected in a same country. A sample made of 76,744 subjects, all living in the same timezone in Brazil, with only one week of difference between questionnaire responses.

# 5.1.2 Results

The local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days (MSF $_{\rm sc}$ ), MCTQ proxy for measuring the chronotype, had an overall mean of 04:28:35. The distribution curve is shown in Figure 5.

That's the midsleep point of Brazilian subjects with an intermediate/average chronotype. One can imagine, following the 7-9h sleep recommendation for healthy adults of the American Academy of Sleep Medicine (AASM) (Watson et al., 2015), that this average person would, if he/she had no social obligations, typically wake up at about 08:28:35.

Figure 5 – Distribution of the local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days (MSF $_{\rm sc}$ ), MCTQ proxy for measuring the chronotype. The categorical cut-offs follow a quantile approach going from extremely early (0| -0.11) to the extremely late (0.88 -1).



Source: Created by the author. Based on data visualization found in Roenneberg, Wirz-Justice, et al. (2019).

The MSF<sub>sc</sub> curve had a skewness of 0.284 and a kurtosis of 2.773. However, the distribution was not normal accordingly to Kolmogorov-Smirnov test (D = 0.03717; p-value = 2e-16) and D'Agostino Skewness test (Z3 = 31.525; p-value = 2.2e-16) (see D'Agostino & Belanger, 1990; also Thode, 2002, 46, p. 101).

A linear regression model was created with MSF $_{\rm sc}$  as the response variable and with age and sex as predictors (R $^2=0.05373$ ; F(2,76741)=2180, p-value = 2e-16), the two most known predictors for chronotype (Roenneberg, Kumar, and Merrow (2007)). A Box-Cox transformation of the response variable was needed to attend to the linear regression model assumptions ( $\lambda=-1.1111$ ; MSF $_{\rm sc}^{\lambda-1}/\lambda$ ). All coefficients were significantly different than 0 (p-value = 2e-16) and, accordingly to D'Agostino Skewness test, the residuals were normal (Z3 = -1.1906; p-value = 0.23383). Residual homoscedasticity was verified by a Score Test for Heteroskedasticity ( $\chi^2=0.00$ ; p-value = 1). No collinearity was found between the predictor variables (variance inflation factor: age = 1.0012; sex = 1.0012).

Another model was created on top of the first one, adding the latitude as a predictor variable (R $^2=0.060698$ ; F(3,76740)=1650, p-value =2e-16). All coefficients were significantly different than 0 (p-value =2e-16) and the residuals were normally distributed accordingly to the D'Agostino Skewness test, (Z3 = 0.0742; p-value = 0.94085). Residual homoscedasticity was verified by a Score Test for Heteroskedasticity ( $\chi^2=0.00$ ; p-value = 1). No collinearity was found between the predictor variables (variance inflation factor: age = 1.0065; sex = 1.0016; latitude = 1.0056). The longitude was not used as a predictor because it presented colinearity with the latitude variable.

An F test for nested models showed a significant reduction of the residual sum of squares (F(1,76740) = 568.94, p-value = 2e-16), meaning that the latitude seems to produce an effect on the chronotype. However, when estimating Cohen's  $f^2$  effect size, the result was negligible (Cohen, 1992) ((0.06069-0.05373)/(1-0.06069)=0.00740).

# 5.1.3 **Discussion**

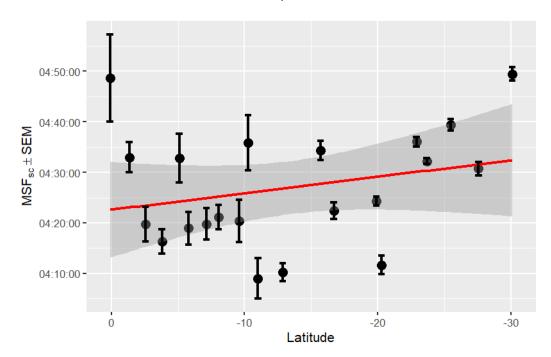
It's important to note that a causal and linear relationship between latitude and chronotype is an *a priori* assumption. The objective of this study is to test or falsify this hypothesis.

For the latitude hypothesis to be true, there must be a significant association between these two variables when the most common covariates are controlled. Predictive models, in this case, are an adequate method for testing this.

The results show that even with a wide latitudinal spectrum and with a big and aligned sample of biological states the latitude effect does not reveal itself in a non-

negligible size. Several studies indicate the existence of this effect on the chronotype (Hut et al., 2013; Leocadio-Miguel et al., 2017; Pittendrigh et al., 1991; Randler, 2008; Randler & Rahafar, 2017; Roenneberg, Wirz-Justice, & Merrow, 2003), but, at this time, at least in humans, no empirical evidence can support this claim. Our results are very similar to Leocadio-Miguel et al. (2017), which also found a negligible effect size (Cohen's  $f^2=0.004143174$ ). The inconsistency of the latitude effect can be visualized in Figure 6.

Figure 6 – Distribution of mean aggregates of the local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days (MSF<sub>sc</sub>), MCTQ proxy for measuring the chronotype, in relation to latitude decimal degree intervals. Higher values of MSF<sub>sc</sub> indicate a tendency toward a late chronotype. The red line represents a linear regression, and the shaded area indicates a pointwise 95% confidence interval.



Source: Created by the author. Based on data visualization found in Leocadio-Miguel et al. (2017).

Despite the lack of evidence, is not uncommon to hear talks insisting that this effect is real and already confirmed. We suspect that this behavior may be derived from a lack of understanding of statistical models and techniques. Although it may be logical and aligned with the overall theory for the evolution of biological temporal systems, it's our role as scientists to eliminate contractions, not pursue them.

The absence of a strong entrainment with the solar zeitgeber shows that the entrainment phenomenon is more complex than we previously imagined. Other hypothe-

ses for the human circadian entrainment, like the entrainment to self-selected light, proposed by Anna Skeldon and Derk-Jan Dijk (2021), need to be tested and may produce significant results. Methods and techiniques for complex systems, like causal loop diagrams and agent-based models (ABM), may help to unravel this phenomenon more properly.

It's important to notice that the results shown here are preliminary. The data still needs some cleaning and to be balanced with Brazil's latest population census. The latitude coordinates used in the analysis are related to subjects' residential state capital and, hence, have low resolution. Even with these results, it may be that a significant latitude effect can still appear at the end of the research.

Despite the several strengths that the dataset used in this study has, it is also important to notice its weaknesses and limitations. The fact that all the subjects were measured in the Spring season is one of them. Since the objective is to catch individuals in different seasonal patterns, the ideal moment to collect this kind of data is in the wintertime, when there is a greater insolation gradient between the equator and the poles. Another one is that this dataset can be influenced by the presence of a Daylight Saving Time (DST) event. This latter issue is explored in more detail in the methods section.

# 5.2 METHODS

# 5.2.1 Measurement instrument

Chronotypes were measured using the core version of the standard Munich ChronoType Questionnaire (MCTQ) (Roenneberg, Wirz-Justice, & Merrow, 2003). MCTQ is a widely validated and widely used self-report questionnaire for measuring the sleep-wake cycle and chronotypes (Roenneberg, Pilz, et al., 2019). It quantifies the chronotype as a state, a biological circadian phenotype, using as a proxy the local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days (MSF<sub>sc</sub>). A sleep correction (SC) is made when a possible sleep compensation related to a lack of sleep on workdays is identified (Roenneberg, 2012).

Subjects were asked to complete an online questionnaire based on the MCTQ Portuguese translation created by Till Roenneberg & Martha Merrow for the EUCLOCK project (Roenneberg & Merrow, 2006) (statements mean cosine distance = 0.921).

They were also asked to provide sociodemographic (e.g., age, sex), geographic (e.g., full residential address), anthropometric (e.g., weight, height), and work/study routine-related data. A deactivated version of the questionnaire can be seen at https://bit.ly/brchrono-form.

# 5.2.2 **Sample**

The sample is made up of 76,744 Brazilian subjects. It was obtained in 2017 from October 15th to 21st by a broadcast of the online questionnaire on a popular Sunday TV show with national reach (*Metade da população se sente mal no horário de verão, revela pesquisa*, 2017). This amount of data collected in such a short time gave the sample a population cross-sectional characteristic.

A survey conducted in 2019 by the Brazilian Institute of Geography and Statistics (IBGE) (2021) found that 82.17% of Brazilian households had access to an internet connection. Therefore, this sample is likely to have a good representation of Brazil's population. Only residents of Brazilian states in the UTC-3 timezone, aged 18 years or older, were included in the final sample.

In order to verify if the sample size was adequate for the study of the phenomenon under investigation, a power analysis was conducted for nested multiple regression models using the G\*Power software (Faul et al., 2007). The analysis used the parameters presented in Leocadio-Miguel et al. (2017) article for a multiple linear regression with 10 tested predictors and only 10 conceived predictors, considering a significance level of 0.05 ( $\alpha$ ) and a power of 0.95 ( $1-\beta$ ). The result showed that a sample of 5,895 individuals would be necessary to test the hypothesis.

Daylight Saving Time (DST) began in Brazil at midnight on November 15th, 2017. Residents from the Midwest, Southeast, and South regions were instructed to set the clock forward by 1 hour. We believe that this event did not contaminate the data since it started on the same day of the data collection. It's important to notice that MCTQ asks subjects to relate their routine behavior, not how they behaved in the last few days. A possible effect of the DST on the sample is the production of an even later chronotype for populations near the planet's poles, amplifying a possible latitude effect. However, this was not shown on the hypothesis test.

Based on the 2022 census (Instituto Brasileiro de Geografia e Estatística, n.d.-a), Brazil had 52.263% of females and 47.737% of males with an age equal to or greater than 18 years old. The sample is skewed for female subjects, with 66.297% of females and 33.703% of male subjects.

The subjects' mean age is 32.015 years (SD = 9.252; Max. = 58.786). Female subjects have a mean age of 31.787 years (SD = 9.364; Max. = 58.786) and male subjects 32.464 years (SD = 9.012; Max. = 58.772). For comparison, based on the 2022 census (Instituto Brasileiro de Geografia e Estatística, n.d.-b), Brazil's population with an age equal to or greater than 18 years old had a mean age of 44.277 years (SD = 17.221), with a mean age of 44.987 years (SD = 17.511) for female subjects and a mean age of 43.499 years (SD = 16.864) for male subjects.

Considering the five regions of Brazil, the sample is mostly skewed for the Southeast, the most populated region. According to Brazil's 2022 census (Instituto Brasileiro de Geografia e Estatística, 2022), the Southeast region is home to 41.784% of Brazil's population, followed by the Northeast (26.910%), South (14.741%), North (8.544%), and Midwest (8.021%) regions. 62.454% of the sample is located in the Southeast region, 11.797% in the Northeast, 17.861% in the South, 1.682% in the North, and 6.205% in the Midwest region. Note that a lack of subjects in the North and Midwest region is justified by the sample timezone inclusion criteria (UTC-3).

The sample latitudinal range was 30.211 decimal degrees (Min. =-30.109; Max. =0.10177) with a longitudinal span of 16.378 decimal degrees (Min. =-51.342; Max. =-34.964). For comparison, Brazil has a latitudinal range of 39.024 decimal degrees (Min. =-33.752; Max. =5.2719) and a longitudinal span of 39.198 decimal degrees (Min. =-34.793; Max. =-73.991).

The results shown in this article are just a preliminary view of the data analysis. The latitudes and longitudes of each subject are represented by the coordinates of his/her state's capital (a low resolution). The final results will have the latitude and longitude coordinates based on subjects' postal codes and will also use a balanced dataset following the latest Brazil census.

# 5.2.3 Analysis

The data wrangling and analysis followed the data science program proposed by Hadley Wickham and Garrett Grolemund (Wickham, 2016). All processes were made with the help of the R programming language (R Core Team, n.d.), RStudio IDE (Posit Team, n.d.), and several R packages. The tidyverse and rOpenSci package ecosystem and other R packages adherents of the tidy tools manifesto (Wickham & Bryan, 2023) were prioritized. The MCTQ data was analyzed using the mctq rOpenSci peer-reviewed package (Vartanian, 2023). All processes were made in order to provide result reproducibility and to be in accordance with the FAIR principles (Wilkinson et al., 2016).

The study hypothesis was tested using nested models of multiple linear regressions. The main idea of nested models is to verify the effect of the inclusion of one or more predictors in the model variance explanation (i.e., the R<sup>2</sup>) (Allen, 1997). This can be made by creating a restricted model and then comparing it with a full model. Hence, the hypothesis can be schematized as follows.

$$\begin{cases} \mathsf{H}_0 : \mathsf{R}^2_{\mathsf{res}} >= \mathsf{R}^2_{\mathsf{full}} \\ \mathsf{H}_a : \mathsf{R}^2_{\mathsf{res}} < \mathsf{R}^2_{\mathsf{full}} \end{cases}$$

In order to test a possible latitude association in predicting the chronotype, the full model was the restricted model with the addition of the latitude variable. The restricted model had the local time of the sleep corrected midpoint between sleep onset and sleep end on work-free days ( $MSF_{sc}$ ) as the response variable, MCTQ proxy for the chronotype, with sex and age as predictors.

A residual analysis was made to ensure the validity of the models before the hypothesis test. The hypothesis was tested using a 0.05 ( $\alpha$ ) significance level.

To favor the alternative hypothesis  $(H_a)$ , not only the  $R^2$  of the full model must be significantly larger than the  $R^2$  of the restricted model, but the effect size must be at least considered small. To evaluate the effect size, Cohen's  $f^2$  and his categorical parameters for size were used (Cohen, 1992). That means that, in order to favor  $(H_a)$ , the effect size must be at least equal to or greater than 0.0219.

No blinding procedures were used during the analysis.

# 5.2.4 Data availability

The data that support the findings of this study are available from the corresponding author [DV]. Restrictions apply to the availability of these data, which were used under the approval of a Research Ethics Committee (REC) linked to the Brazilian National Research Ethics Committee (CONEP), hence it cannot be publicly shared. Data are, however, available from the author upon reasonable request and with CONEP approval.

# 5.2.5 Code availability

The research compendium of the project is available under the MIT license at https://github.com/danielvartan/mastersthesis. The code has all the steps from the raw data to the test results.

# 5.3 ACKNOWLEDGMENTS

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# 5.4 ETHICS DECLARATIONS

# 5.4.1 Competing interests

The author declares that the study was carried out without any commercial or financial connections that could be seen as a possible competing interest.

# 5.5 ADDITIONAL INFORMATION

This manuscript shows only preliminary results and should not be considered as a document ready for journal submission.

See the appendices section for supplementary information.

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# 5.6 RIGHTS AND PERMISSIONS

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# 6 CONCLUSION

# ! Important

You are reading the work-in-progress of this thesis.

This chapter is currently a dumping ground for ideas, and I don't recommend reading it.

#### **REFERENCES\***

- Allen, M. P. (1997). Understanding regression analysis. Plenum Press.
- Andrade, T. G. de, & Beale, A. D. (2024). Darwin and the biological rhythms. *PNAS Nexus*, *3*(8), pgae318. https://doi.org/10.1093/pnasnexus/pgae318
- Aschoff, J. (Ed.). (1981). *Biological rhythms* (Vol. 4). Plenum Press. https://doi.org/10.1007/978-1-4615-6552-9
- Aschoff, J. (1989). Temporal orientation: Circadian clocks in animals and humans. *Animal Behaviour*, 37, 881–896. https://doi.org/10.1016/0003-3472(89)90132-2
- Boccara, N. (2010). *Modeling complex systems* (2nd ed.). Springer New York. https://doi.org/10.1007/978-1-4419-6562-2
- Borbély, A. A. (1982). A two process model of sleep regulation. *Human Neurobiology*, *1*(3), 195–204. https://pubmed.ncbi.nlm.nih.gov/7185792/
- Borbély, A. A., Daan, S., Wirz-Justice, A., & Deboer, T. (2016). The two-process model of sleep regulation: A reappraisal. *Journal of Sleep Research*, 25(2), 131–143. https://doi.org/10.1111/jsr.12371
- Cambridge University Press. (n.d.). *Cambridge dictionary*. Retrieved September 21, 2023, from https://dictionary.cambridge.org/
- Cohen, J. (1992). A power primer. *Psychological Bulletin*, *112*(1), 155–159. https://doi.org/10.1037/003 3-2909.112.1.155
- D'Agostino, R. B., & Belanger, A. (1990). A suggestion for using powerful and informative tests of normality. *The American Statistician*, 44(4), 316–321. https://doi.org/10.2307/2684359
- de Mairan, J.-J. (1729). Observation botanique. Histoire de l'Académie royale des sciences avec les mémoires de mathématique et de physique tirés des registres de cette Académie, 35. Retrieved October 17, 2024, from http://www.bibnum.education.fr/sciencesdelavie/biologie/observation-botani que
- Ehret, C. F. (1974). The sense of time: Evidence for its molecular basis in the eukaryotic gene-action system. In *Advances in Biological and Medical Physics* (pp. 47–77, Vol. 15). Elsevier. https://doi.org/10.1016/B978-0-12-005215-8.50009-7
- Faul, F., Erdfelder, E., Lang, A.-G., & Buchner, A. (2007). G\*Power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods*, 39(2), 175–191. https://doi.org/10.3758/BF03193146
- Foster, R. G., & Kreitzman, L. (2005). Rhythms of life: The biological clocks that control the daily lives of every living thing. Profile Books.
- Frommlet, F., Bogdan, M., & Ramsey, D. (2016). *Phenotype and genotype: The search for influential genes* (Vol. 18). Springer London. https://doi.org/10.1007/978-1-4471-5310-8
- Horne, J. A., & Ostberg, O. (1976). A self-assessment questionnaire to determine morningness-eveningness in human circadian rhythms. *International Journal of Chronobiology*, *4*(2), 97–110.
- Horzum, M. B., Randler, C., Masal, E., Beşoluk, Ş., Önder, İ., & Vollmer, C. (2015). Morningness–eveningness and the environment hypothesis a cross-cultural comparison of Turkish and German adolescents. *Chronobiology International*, 32(6), 814–821. https://doi.org/10.3109/07420528.2015.1041598
- Hut, R. A., Paolucci, S., Dor, R., Kyriacou, C. P., & Daan, S. (2013). Latitudinal clines: An evolutionary view on biological rhythms. *Proceedings of the Royal Society B: Biological Sciences*, *280*(1765), 20130433. https://doi.org/10.1098/rspb.2013.0433

<sup>\*</sup> In accordance with the American Psychological Association (APA) Style, 7th edition.

- Instituto Brasileiro de Geografia e Estatística. (n.d.-a). *Tabela 1378 população residente, por situação do domicílio, sexo e idade, segundo a condição no domicílio e compartilhamento da responsabilidade pelo domicílio* (Tabela). Tabela. SIDRA. https://sidra.ibge.gov.br/tabela/1378
- Instituto Brasileiro de Geografia e Estatística. (n.d.-b). *Tabela 9514: População residente, por sexo, idade e forma de declaração da idade* (Tabela). Tabela. SIDRA. Retrieved November 11, 2023, from https://sidra.ibge.gov.br/tabela/9514
- Instituto Brasileiro de Geografia e Estatística. (2021). Pesquisa nacional por amostra de domicílios contínua: Acesso à internet e à televisão e posse de telefone móvel celular para uso pessoal 2019. Instituto Brasileiro de Geografia e Estatística. Rio de Janeiro, RJ. https://biblioteca.ibge.gov.br/visua lizacao/livros/liv101794\_informativo.pdf
- Instituto Brasileiro de Geografia e Estatística. (2022). Censo demográfico 2022: População por idade e sexo: Resultados do universo. IBGE. Rio de Janeiro, RJ. Retrieved November 11, 2023, from https://biblioteca.ibge.gov.br/index.php/biblioteca-catalogo?view=detalhes&id=73102
- Khalsa, S. B. S., Jewett, M. E., Cajochen, C., & Czeisler, C. A. (2003). A phase response curve to single bright light pulses in human subjects. *The Journal of Physiology*, *549*(3), 945–952. https://doi.org/10.1113/jphysiol.2003.040477
- Kuhlman, S. J., Craig, L. M., & Duffy, J. F. (2018). Introduction to chronobiology. *Cold Spring Harbor Perspectives in Biology*, *10*(9), a033613. https://doi.org/10.1101/cshperspect.a033613
- Laboratory, C. S. H. (n.d.). 1960: Biological clocks, vol. XXV. Retrieved July 17, 2023, from https://symposium.cshlp.org/site/misc/topic25.xhtml
- Latinitium. (n.d.). *Latin dictionaries*. Latinitium. Retrieved September 21, 2023, from https://latinitium.com/latin-dictionaries/
- Leocadio-Miguel, M. A., Louzada, F. M., Duarte, L. L., Areas, R. P., Alam, M., Freire, M. V., Fontenele-Araujo, J., Menna-Barreto, L., & Pedrazzoli, M. (2017). Latitudinal cline of chronotype. *Scientific Reports*, 7(1), 5437. https://doi.org/10.1038/s41598-017-05797-w
- Leocadio-Miguel, M. A., Oliveira, V. C. D., Pereira, D., & Pedrazzoli, M. (2014). Detecting chronotype differences associated to latitude: A comparison between Horne-Östberg and Munich Chronotype questionnaires. *Annals of Human Biology*, *41*(2), 107–110. https://doi.org/10.3109/03014460.2013.8 32795
- Marques, N., & Menna-Barreto, L. (Eds.). (2003). *Cronobiologia: Princípios e aplicações* (3. ed. rev. e ampl.). Editora da Universidade de São Paulo.
- Menna-Barreto, L. (2003). O tempo na biologia. In *Cronobiologia: Princípios e aplicações* (3. ed. rev. e ampl., pp. 25–30). Editora da Universidade de São Paulo.
- Menna-Barreto, L., & Marques, N. (Eds.). (2023, August 16). *História e perspectivas da cronobiologia no brasil e na américa latina*. Editora da Universidade de São Paulo.
- Metade da população se sente mal no horário de verão, revela pesquisa [tvbroadcast]. (2017, October 15). Rio de Janeiro, RJ, Rede Globo. https://globoplay.globo.com/v/6219513/
- Minors, D. S., Waterhouse, J. M., & Wirz-Justice, A. (1991). A human phase-response curve to light. *Neuroscience Letters*, 133(1), 36–40. https://doi.org/10.1016/0304-3940(91)90051-T
- Nobel Prize Outreach AB. (n.d.). *Press release*. The Nobel Prize. Retrieved September 28, 2023, from https://www.nobelprize.org/prizes/medicine/2017/press-release/
- Paranjpe, D. A., & Sharma, V. K. (2005). Evolution of temporal order in living organisms. *Journal of Circadian Rhythms*, 3. https://doi.org/10.1186/1740-3391-3-7
- Pittendrigh, C. S. (1981). Circadian systems: General perspective. In *Biological rhythms* (pp. 57–80, Vol. 4). Plenum Press. https://doi.org/10.1007/978-1-4615-6552-9
- Pittendrigh, C. S. (1993). Temporal organization: Reflections of a darwinian clock-watcher. *Annual Review of Physiology*, 55(1), 17–54. https://doi.org/10.1146/annurev.ph.55.030193.000313

- Pittendrigh, C. S., Kyner, W. T., & Takamura, T. (1991). The amplitude of circadian oscillations: Temperature dependence, latitudinal clines, and the photoperiodic time measurement. *Journal of Biological Rhythms*, 6(4), 299–313. https://doi.org/10.1177/074873049100600402
- Posit Team. (n.d.). RStudio: Integrated development environment for R. http://www.posit.co
- R Core Team. (n.d.). R: A language and environment for statistical computing. Vienna, Austria. https://www.R-project.org
- Randler, C. (2008). Morningness □ eveningness comparison in adolescents from different countries around the world. *Chronobiology International*, *25*(6), 1017–1028. https://doi.org/10.1080/07420520 802551519
- Randler, C., & Rahafar, A. (2017). Latitude affects morningness-eveningness: Evidence for the environment hypothesis based on a systematic review. *Scientific Reports*, 7(1), 39976. https://doi.org/10.10 38/srep39976
- Roenneberg, T. (2012). What is chronotype? *Sleep and Biological Rhythms*, *10*(2), 75–76. https://doi.org/10.1111/j.1479-8425.2012.00541.x
- Roenneberg, T., Allebrandt, K. V., Merrow, M., & Vetter, C. (2012). Social jetlag and obesity. *Current Biology*, 22(10), 939–943. https://doi.org/10.1016/j.cub.2012.03.038
- Roenneberg, T., Daan, S., & Merrow, M. (2003). The art of entrainment. *Journal of Biological Rhythms*, 18(3), 183–194. https://doi.org/10.1177/0748730403018003001
- Roenneberg, T., Hut, R., Daan, S., & Merrow, M. (2010). Entrainment concepts revisited. *Journal of Biological Rhythms*, 25(5), 329–339. https://doi.org/10.1177/0748730410379082
- Roenneberg, T., Kuehnle, T., Juda, M., Kantermann, T., Allebrandt, K., Gordijn, M., & Merrow, M. (2007). Epidemiology of the human circadian clock. *Sleep Medicine Reviews*, *11*(6), 429–438. https://doi.org/10.1016/j.smrv.2007.07.005
- Roenneberg, T., Kumar, C. J., & Merrow, M. (2007). The human circadian clock entrains to sun time. *Current Biology*, 17(2), R44–R45. https://doi.org/10.1016/j.cub.2006.12.011
- Roenneberg, T., & Merrow, M. (2006). *EUCLOCK: Portuguese MCTQ*. Retrieved July 17, 2023, from http://web.archive.org/web/20141115175303/https://www.bioinfo.mpg.de/mctq/core\_work\_life/core/core.jsp?language=por\_b
- Roenneberg, T., & Merrow, M. (2016). The circadian clock and human health. *Current Biology*, 26(10), R432–R443. https://doi.org/10.1016/j.cub.2016.04.011
- Roenneberg, T., Pilz, L. K., Zerbini, G., & Winnebeck, E. C. (2019). Chronotype and social jetlag: A (self-) critical review. *Biology*, 8(3), 54. https://doi.org/10.3390/biology8030054
- Roenneberg, T., Wirz-Justice, A., & Merrow, M. (2003). Life between clocks: Daily temporal patterns of human chronotypes. *Journal of Biological Rhythms*, *18*(1), 80–90. https://doi.org/10.1177/07487304 02239679
- Roenneberg, T., Wirz-Justice, A., Skene, D. J., Ancoli-Israel, S., Wright, K. P., Dijk, D.-J., Zee, P., Gorman, M. R., Winnebeck, E. C., & Klerman, E. B. (2019). Why should we abolish daylight saving time? Journal of Biological Rhythms, 34(3), 227–230. https://doi.org/10.1177/0748730419854197
- Rotenberg, L., Marques, N., & Menna-Barreto, L. (2003). História e perspectivas da cronobiologia. In *Cronobiologia: Princípios e aplicações* (3. ed. rev. e ampl., pp. 31–53). Editora da Universidade de São Paulo.
- Skeldon, A. C., & Dijk, D.-J. (2021). Weekly and seasonal variation in the circadian melatonin rhythm in humans: Entrained to local clock time, social time, light exposure or sun time? *Journal of Pineal Research*, 71(1), e12746. https://doi.org/10.1111/jpi.12746
- Thode, H. C. (2002). Testing for normality. Marcel Dekker.
- Vartanian, D. (2023). {Abnt}: Quarto format designed for theses and dissertations that adhere to the standards of the Brazilian Association of Technical Standards (ABNT) (Version 0.1.1). https://github.com/danielvartan/abnt

- Watson, N. F., Badr, M. S., Belenky, G., Bliwise, D. L., Buxton, O. M., Buysse, D., Dinges, D. F., Gangwisch, J., Grandner, M. A., Kushida, C., Malhotra, R. K., Martin, J. L., Patel, S. R., Quan, S. F., & Tasali, E. (2015). Recommended amount of sleep for a healthy adult: A joint consensus statement of the American Academy of Sleep Medicine and Sleep Research Society. *Journal of Clinical Sleep Medicine*, *11*(6), 591–592. https://doi.org/10.5664/jcsm.4758
- Wickham, H. (2016). *Ggplot2: Elegant graphics for data analysis* (2nd ed.). Springer International Publishing. https://doi.org/10.1007/978-3-319-24277-4
- Wickham, H., & Bryan, J. (2023). *R packages* (2nd ed.). O'Reilly Media. Retrieved July 7, 2024, from https://r-pkgs.org/
- Wilkinson, M. D., Dumontier, M., Aalbersberg, IJ. J., Appleton, G., Axton, M., Baak, A., Blomberg, N., Boiten, J.-W., Da Silva Santos, L. B., Bourne, P. E., Bouwman, J., Brookes, A. J., Clark, T., Crosas, M., Dillo, I., Dumon, O., Edmunds, S., Evelo, C. T., Finkers, R., ... Mons, B. (2016). The FAIR guiding principles for scientific data management and stewardship. *Scientific Data*, 3(1), 160018. https://doi.org/10.1038/sdata.2016.18